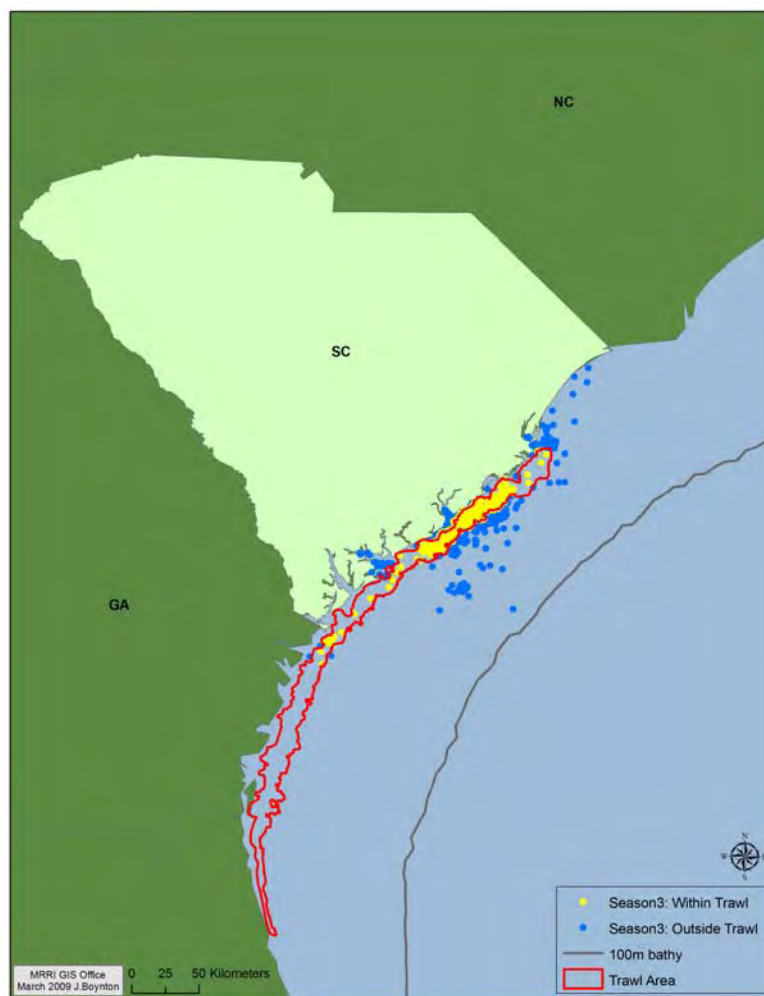


# **Examination of Local Movement and Migratory Behavior of Sea Turtles During Spring and Summer Along the Atlantic Coast Off the Southeastern United States**

## **Final Project Report To The National Marine Fisheries Service National Oceanic and Atmospheric Administration**



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# **FINAL REPORT TO NATIONAL MARINE FISHERIES SERVICE**

For

Examination of Local Movement and Migratory Behavior of Sea Turtles During Spring and  
Summer Along the Atlantic Coast Off the Southeastern United States

by

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## Executive Summary

In response to low loggerhead re-encounter rates (15 of 945 total collections) during 2000-2003, as well as infrequent collection of loggerheads ( $n=11$ ) tagged by other programs during the same period, satellite telemetry studies were initiated with juvenile loggerheads collected near Charleston, SC, to document seasonal distributional patterns for which only sparse historical data were available. Eighty-five percent of 34 satellite tagged juveniles remained resident off the coasts of SC and GA between April and November, when water temperatures were above 17°C. Sixty-two percent of daily locations were estimated to occur within the boundaries of the regional trawl survey area sampled during 2000-2003. Loggerheads predominantly moved further offshore during the winter and returned to the same coastal areas occupied the previous spring through fall. Data demonstrated high probability of occurrence within the regional trawl survey area during the June-July sampling period. A generally resident nature, but with highly variable levels of mobility, may also contribute greatly to low re-encounter rates within our survey as well as low probability of being recaptured by coastal surveys other than our own.

In April 2006 and 2007, additional efforts focused on collection of adult male loggerheads aggregated for mating in the Port Canaveral, FL, shipping entrance channel. Study objectives included assessment of reproductive condition, methodology comparison for assessing reproductive condition, and characterization of temporal and spatial distribution patterns via satellite telemetry. Ninety percent of adult male loggerheads collected from the Port Canaveral, FL, shipping entrance channel were reproductively active. More than half of adult male loggerheads collected were transient animals, which were slightly larger than resident males. All transient males were reproductively active, and most resident males were also active. Residents and transients co-occurred in near shore waters during April and mid-May, after which time residents moved offshore to deeper waters (>26m) and transients dispersed to multiple locations along the U.S. East Coast, the northern Gulf of Mexico, and the FL Keys/Bahamas.

Fishery-independent trawling in the regional trawl survey area was resumed in 2008 following a five-year hiatus. Overall loggerhead catch rates in 2008 were 1.5 times greater than in 2000; however, significant differences were not detected between 2000-2003 and 2008. Significantly greater CPUE was detected for two loggerhead size classes, representing loggerheads that are maturing or are already mature (75.1 to 85.0cm SCLmin) and loggerheads 'next in line' to become mature (65.1 to 75.0cm SCLmin). Increased catch rates can not automatically be assumed to represent increased abundance due to the inability to assess the probability of loggerhead detection at the time of sampling; however, increased catch rates in the regional survey since 2000 are 43 times greater than in fishery-dependent coastal surveys conducted prior to 1976 and 14-23 times greater than in fishery-dependent and -independent coastal surveys conducted in between the mid-1970's and the early 1990's; thus, it is highly plausible that inherent increases in population have indeed occurred, even if they can't be precisely measured.

Loggerhead catch rates in the Charleston channel were greater than in the early 1990's, and a discernable increase in size distribution between the two study periods suggests growth among strong year classes that continue to persist. Growth within slightly smaller size classes collected elsewhere also suggests stable to strong loggerhead recruitment, despite a decline in the smallest (and least likely to be encountered) loggerheads collected by our various efforts since 2000.

## Introduction

Loggerhead sea turtles (*Caretta caretta*) are the most commonly occurring sea turtle species in coastal waters along the Southeastern United States (SE USA) and represent the progeny of multiple rookeries (Bowen et al., 1993; Sears et al., 1995; TEWG, 2000; Maier et al., 2004). Tagging studies of nesting female loggerheads suggest that most return to the same beaches in successive breeding seasons (Bjorndal et al., 1983) and it is widely accepted that most females return to their natal regions to nest. Although considerable effort has been expended to study adult females on nesting beaches, much less is known about the seasonal distributional patterns of juveniles and adult males in coastal waters; hence, the importance of conducting in-water studies with sea turtles to complement nesting and stranding data.

Prior to May 2000, in-water studies targeting sea turtles were primarily conducted at shipping entrance channels (Butler et al., 1987; Standora et al., 1993a; Dickerson et al., 1995) or at opportunistic inshore collection areas such as where pound nets were located (Byles, 1988; Epperly et al., 1995a; Morreale and Standora, 1994). The need to conduct, "...long-term, in-water indices of loggerhead abundance in coastal waters" (TEWG, 1998) led to the development of a regional in-water survey of loggerheads during summers 2000-2003 (Maier et al., 2004). Coastal waters 1-15km offshore between Winyah Bay, SC, and St. Augustine, FL, were sampled in late spring and summer in a nearly simultaneous manner using three research vessels. High catch rates were reported (Maier et al., 2004); however, very low recapture rates (<2%) were also reported, the cause of which was not readily evident.

In an effort to better understand the potential influence of seasonal distributional patterns of juvenile loggerheads on regional trawl survey tag-recapture rates, the focus of the in-water survey was modified beginning in 2004 to intensively target one small trawling area to: (1) examine the effect of intensive trawling on recapture rates and (2) quickly obtain an adequate sample size of turtles to outfit with satellite transmitters. Prior to this study, satellite telemetry had only been attempted with three juvenile loggerheads collected south of Cape Hatteras (NMFS, unpublished data 1; USACOE, unpublished data); thus, long-term information on habitat utilization of juveniles in coastal waters was virtually non-existent for this region. In order to facilitate historical comparisons of catch-per-unit effort (Van Dolah and Maier, 1993; Dickerson et al., 1995), the shipping entrance channel of Charleston harbor was selected for this trawl survey. Logistical considerations, including close proximity to a turtle rehabilitation facility at the SC Aquarium in Charleston, also contributed to the decision to restrict trawling to this location. Between May 2004 and August 2007, 34 non-rehabilitated juvenile loggerheads were satellite-tagged for this study.

During April 2006 and 2007, a second trawling area (the Port Canaveral, FL, shipping entrance channel) was added to this study to facilitate collection of adult male loggerheads during their presumed mating aggregation in close proximity to the most productive loggerhead nesting beaches along the U.S. Eastern Seaboard. The purpose of this research was two-fold. First, to utilize new techniques to refine the ability assess reproductive biology and physiology of adult male loggerheads, which had only been studied (Wibbels et al., 1987) once prior and nearly 30 years earlier at this very important breeding location. Similarly, the second goal of this research was to utilize satellite-telemetry to investigate the temporal and spatial distributional patterns of

adult male loggerheads, for both reproductively active and inactive individuals. Prior to commencing this work, satellite-telemetry studies with adult males along the U.S. Eastern Seaboard had only been conducted with two adult males collected from Chesapeake Bay (Keinath, 1993) and five adult males collected from Florida Bay (NMFS, unpublished data 2). Furthermore, only six (of 120) loggerheads tagged with acoustic and/or radio transmitters near Canaveral since 1981 were adult males; thus, long-term data sets for adult males were virtually non-existent for northern subpopulation as well as the very important Canaveral study area.

Following a five-year hiatus as well as successful completion of four years of data collection for juvenile loggerheads and two years of data collection for adult males, the focus of the in-water turtle trawl survey reverted to its original randomized sampling design in summer 2008. The purpose of resuming the regional survey was primarily to document potential changes in catch-per-unit-effort and size frequency distributions, to better understand potential population responses of loggerheads to a plethora of management policies which have been implemented since 1978. In addition to standard sea turtle catch demographic data, blood and tissue samples were collected from loggerhead sea turtles to assess a variety of health parameters. Non-turtle by-catch assessments were also conducted, in an attempt to determine if spatial distribution of turtle catch is influenced by prey species distributions, as well as to assess what impacts, if any, conducting an intensive trawl survey for sea turtles has on sensitive benthic habitats.

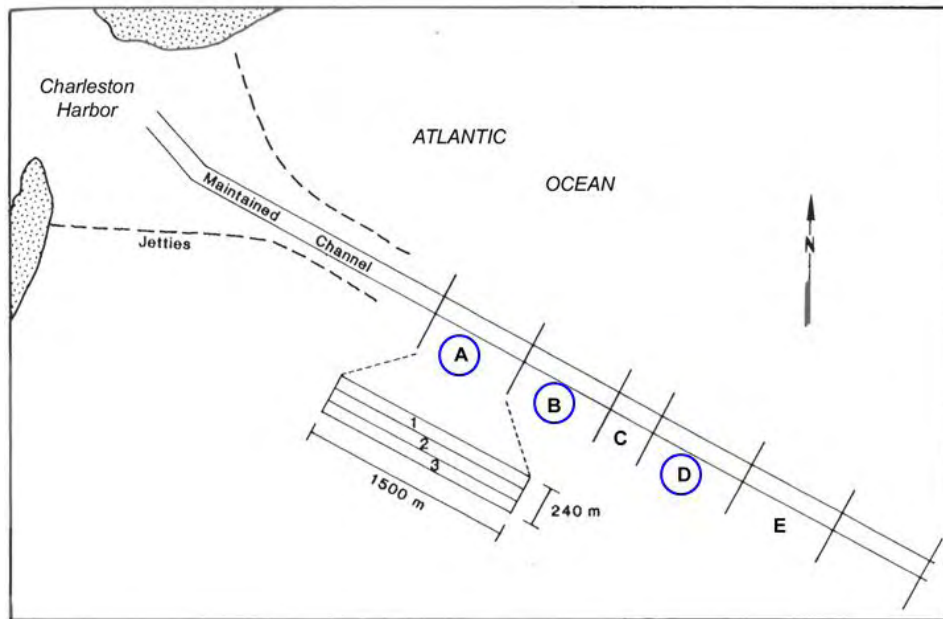
Detailed tracking maps for juvenile and adult male loggerheads have been presented in Annual grant reports. This final report summarizes key aspects of our findings, with greater emphasis on statistical analyses than has been presented previously. Subsequently, the results section of this report is organized as a series of chapters, which have predominantly been written in manuscript format to facilitate expeditious efforts to publish these important findings later this year.

## **Methods**

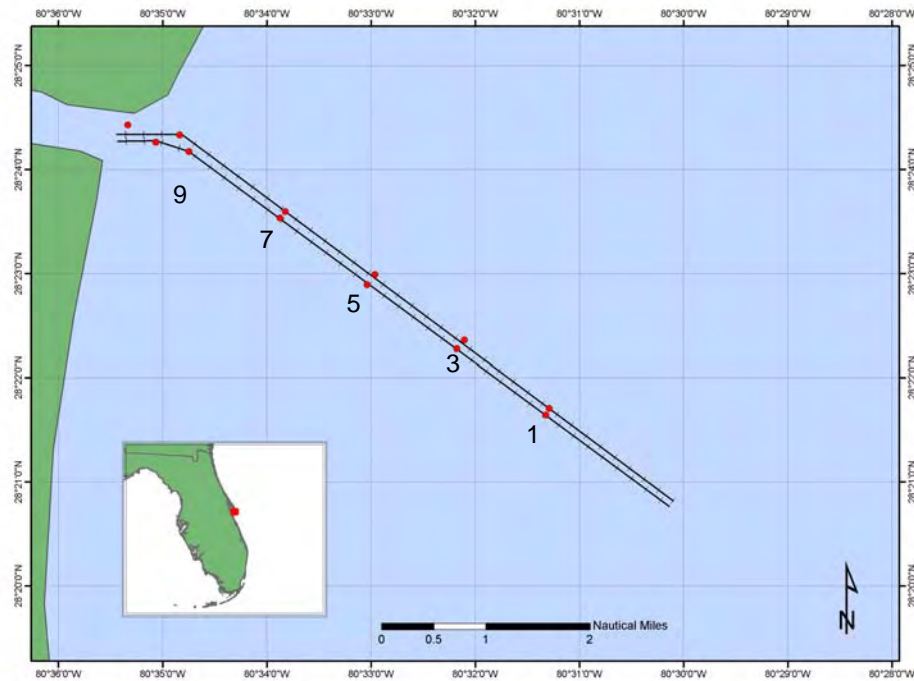
### *Study Areas, Research Vessels, and Trawl Specifications*

Trawling in the Charleston, SC, shipping entrance channel (32°42'N, -79°48'W; Figure 1) was conducted between channel markers "17/18" and "13/14". Seven of 12 index stations first utilized in 1990-1991 (Van Dolah and Maier, 1993) were sampled for this research; stations in the "E" block and in the center of the channel (A-2, B-2, D-2) were dropped following unsuccessful (due to rough bottom) attempts to sample these stations with trawls in May 2004. Sampling was conducted in 2004 (May, June and August 2004), 2005 (May and August), 2006 (May) and 2007 (May and August). Stations were systematically trawled during 2004-2006; however, stations with high catch rates were targeted in 2007 in order to expedite sea turtle collection. Trawl bottom time in the Charleston shipping channel ranged from 9 to 21 minutes.

Trawling was conducted between channel markers "1/2" and "9/10" in the shipping entrance channel to Port Canaveral, FL (28°23'N, -80°32'W; Figure 2). A total of three, five-day cruises were conducted in April 2006 (one cruise) and April 2007 (two cruises). Fifteen minute trawls (bottom time) were conducted between subsequent channel markers (1 to 3; 3 to 5; etc.). Due to the principal objective of collecting adult male loggerheads as quickly as possible, opportunistic (rather than randomized) sampling was employed.



**Figure 1.** Index trawling blocks (Van Dolah and Maier, 1993) in the Charleston, SC, shipping entrance channel sampled in 2004-2007 (indicated by blue circles).



**Figure 2.** Index trawling blocks between navigational buoys (numbers shown) in the Port Canaveral, FL, shipping entrance channel.



Trawling in 2008 was conducted at randomly selected stations in coastal waters (4.6 to 12.2 m) between Winyah Bay, SC, and St. Augustine, FL (Figure 3). The R/V *Georgia Bulldog* sampled south of Savannah, GA, and the R/V *Lady Lisa* sampled north of Savannah, GA. A coin toss determined which direction the first cruise for each vessel would start relative to their homeport, and weekly direction was systematically alternated thereafter. Near shore (<1 to 5km) and further offshore (5 to 12km) stations were both sampled before and after noon to prevent fine scale spatial-temporal biases. Per permit modifications, trawl duration was 20 minutes (bottom time) which represented a 33% reduction in sampling effort relative to 2000-2003.



**Figure 3.** Regional trawl survey area between Winyah Bay, SC, and St. Augustine, FL.

All trawl sampling between 2004 and 2008 was conducted aboard double-rigged shrimp trawlers (R/V *Georgia Bulldog*, 72', and the R/V *Lady Lisa*, 75') towing at speeds of 2.5-3.0 knots. Sampling in the Charleston, SC, shipping entrance channel was conducted by the R/V *Georgia Bulldog* in May 2004; however, the R/V *Lady Lisa* completed all other sampling at this location. All trawl sampling in the Port Canaveral, FL, shipping entrance channel was completed by the R/V *Georgia Bulldog*. Lastly, sampling during the regional survey in 2008 was completed by both research vessels, with the R/V *Lady Lisa* operating from the Savannah, GA, shipping entrance channel to Winyah Bay, SC, and the R/V *Georgia Bulldog* operating from the Savannah, GA, shipping entrance channel to St. Augustine, FL. Fiscal constraints necessitated scaling down the trawling operation from three (2000-2003) to two research vessels, which further reduced sampling effort in 2008 relative to 2000-2003; however, a priori boot-strap analyses using 2000-2003 data demonstrated that the ability to make inter-annual comparisons would not be adversely affected by the proposed reduction in annual sampling effort.

Standardized National Marine Fisheries Service (NMFS) turtle nets were utilized for this study. Turtle nets were paired 60-foot (head-rope), 4-seam, 4-legged, 2-bridal nets. Net body consisted of 4" bar and 8" stretch mesh, with top's sides made of #36 twisted and bottom consisting of #84 braided nylon line. Cod end (60' corkline to cod end) consisted of 2" bar and 4" stretch mesh.

### *Capture and General Processing*

Turtles were immediately removed from nets and examined for life-threatening injuries, before being visually/electronically scanned for existing tags. If not previously tagged in this study, a sequential project identification number was assigned to each turtle.

Blood samples were collected for all sea turtles >5kg body weight with a 21ga, 1.5 in. needle from the dorsal cervical sinus of loggerhead turtles as described by Owens and Ruiz (1980). Blood samples consisted of a maximum of 45ml total volume and did not exceed the total recommended volume (10% of total blood volume) based upon total weight as described by Jacobson (1998). Blood samples were collected for the following collaborators and purposes:

genetics - 5ml (University of South Carolina & University of Georgia)

sex determination - 10ml (College of Charleston)

CBC/Blood chemistry -- 3ml (Antech Diagnostics)

Toxicological screening and immunological bioassay – 20ml (National Institute of Standards and Technology; Medical University of SC)

A suite of standard (Bolten, 1999) morphometric measurements were collected for all sea turtle species sampled. Six straight-line measurements (cm) were made using tree calipers for minimum (CLmin) and notch-tip (CLnt) carapace length, carapace width (CW), head width (HW) and body depth (BD). Curved measurements of CLmin, CLnt and CW were recorded using a nylon tape measure. Additional curved measurements included plastron width (PW), tip of plastron to tip of tail (PT) and tip of cloaca to tip of tail (CT)). Turtles were placed in a nylon mesh harness, slowly raised off the deck, and body weight (kg) recorded using spring scales.

All sea turtles >5kg received two Inconel flipper tags and one Passive Integrated Transponder (PIT) tag (Biomark, Inc.). Triple tagging minimized the probability of complete tag loss. Inconel flipper tags were provided by the Cooperate Marine Turtle Tagging Program (CMTTP). Per the instructions provided by the CMTTP, tags were cleaned to remove oil and residue prior to application. Inconel tag insertion sites, located between the first and second scales on the trailing edge of the front flippers, were swabbed with betadine prior to tag application to create a more aseptic environment. PIT tag insertion points, located in the right front shoulder near the base of the flipper, were also swabbed with betadine prior to the intramuscular injection of the sterile-packed PIT tag. Prior to releasing turtles, a digital photograph of each turtle in a standard 'pose' (dorsal surface exposed, orientation from anterior to posterior) was recorded. Additional photographs of unusual markings or injuries were also recorded.

Additional blood and tissue samples collected for collaborators, as well as by-catch work-up procedures, are described in the methods sections for their respective chapters.

### *Data management*

Hard copy data were recorded on various forms at sea; electronically keyed into a MS Access database at the end of the cruise season; and proofed for errors. In 2008, data were entered electronically at sea using laptop computers, allowing early detection and correction of errors.

## Chapter 1 Sea turtle catch demographics from multi-year coastal trawl surveys in South Carolina, Georgia and northern Florida.

### Introduction

Species composition of sea turtles on foraging grounds is influenced by geographic distributions and habitat preferences. Of the seven extant sea turtle species, five are indigenous to coastal and inland waters of the southern and eastern United States. Along the U.S. East Coast, loggerhead (*Caretta caretta*) and Kemp's ridley (*Lepidochelys kempii*) sea turtles are ubiquitously distributed from estuarine waters to across the continental shelf (Lutcavage and Musick, 1985), and both species have been found as far north as Canadian waters (Squires, 1954; Bleakney, 1955). Green sea turtles (*Chelonia mydas*) are more tropically distributed; however, juvenile green turtles have also been reported from Canadian waters on the U.S. Western Seaboard (McAlpine et al., 2004). Green sea turtles may concentrate close to shore (Makowski et al., 2006), reflecting their herbivorous diet (Arthur et al., 2008) and subsequent restriction to water depths which support photosynthesis. Leatherback sea turtles (*Dermochelys coriacea*) feed on gelatinous zooplankton and undergo long-distance migrations in pursuit of prey (Hays et al., 2006); thus, distribution of leatherbacks in coastal waters is most likely ephemeral rather than sustained. Circum-tropically distributed hawksbill sea turtles (*Eretmochelys imbricata*) forage on reefs and associated fauna (Carr et al., 1966; Meylan, 1988), and accounts of hawksbills north of central Florida are rare.

Genetic studies suggest natal homing to a region of origin among adult females (Bowen and Karl, 2007); wandering and gene flow among adult males (Bowen et al., 2005; Fitzsimmons et al., 1997); and increasing site fidelity with size/age among juveniles on foraging grounds (Laurent et al., 1998; Bowen et al., 2004, 2005). Loggerheads nest on both sides of the Atlantic Ocean and throughout the Gulf of Mexico and Caribbean Sea, and loggerheads collected from the northern foraging ground (FL to Northeast US) are reported to have originated from a number of nesting colonies (Encalada et al., 1998; Bowen et al., 2004). Nesting for Kemp's ridley sea turtles occurs primarily in a narrow corridor in the western Gulf of Mexico; however, isolated nesting is also documented in SC and GA (PINS, 2005; SCDNR). Nesting of green and leatherback sea turtles in SC and GA is rare (SCDNR; GADNR); however, the FL green turtle nesting aggregation is "recognized as a regionally significant colony (USFWS)", and nesting for both green and leatherback sea turtles in FL has increased steadily since 1989 (FFWCC).

Region of origin may influence metrics measured among sea turtles encountered on foraging grounds. Spatial and temporal differences in sex ratios could represent disproportionate contribution of individuals from nesting colonies with female- or male-biases. Large differences in size distributions among foraging grounds likely represent ontogenetic changes in habitat requirements; however, subtle size differences could also reflect the nutritional quality of the foraging ground, energetic costs to reach the foraging ground, recruitment success and/or genetic pre-disposition for growth (which would also render size a less reliable proxy for age). The objective of this chapter is to present size, sex and genetic distribution data for sea turtles collected between 2004 and 2008, with comparisons to sea turtles collected between 2000 and 2003 (Maier et al., 2004) where appropriate.

## Methods

### *Sampling locations*

Between 2004 and 2008, sea turtles were collected by trawling in three temporally and spatially distinct sub-studies, two of which were conducted in shipping entrance channels. Trawling in the both shipping entrance channels was conducted in the manner described in the General Methods section of this report. Catch per unit effort and results of satellite telemetry for loggerheads collected from both locations are presented in Chapters 3-5. Evaluation of methods for assessing reproductive condition is published (Blanvillain et al., 2008).

Randomized trawling in coastal waters (4.6 to 12.2m) between Winyah Bay, SC, and St. Augustine, FL, was conducted between late May and late July 2008; however, loggerhead “hot spots” were re-sampled in August 2008 in an attempt to recapture tagged turtles for clinical studies. The regional sampling area in 2008 was unchanged from the regional sampling area of 2000-2003. For the regional sampling area, four sub-regions were designated based on strata codes used by the Southeastern Area Monitoring and Assessment Program (SEAMAP). The northern portion of strata 27-28 through strata 33-34 represented St. Augustine, FL, to Brunswick, GA. Strata 35-36 to 39-40 approximated Brunswick, GA, to Savannah, GA. Strata 41-42 to 45-46 corresponded to Savannah, GA, to Charleston, SC. And strata 47-48 and 49-50 were designated as Charleston, SC, to Winyah Bay, SC.

Data from a fourth sub-study (fishery-dependent sampling near Charleston, SC, during June 2000-2003) were also included for comparative purposes, given that demographic data were not detailed in a previous contract report (Maier et al., 2004). Fishery-dependent sampling near Charleston, SC, occurred between latitudes 32.650°N and 32.850 °N. Fishery-dependent sampling near Brunswick, GA, was only conducted in June 2000.

### *Turtle measurements and blood sample collection*

Morphometric measurements of sea turtles and blood samples were collected and processed as described in the General Methods section of this report. Straight-line carapace length measurements for adult male loggerheads collected from the Port Canaveral, FL, channel in April 1992 (Dickerson et al., 1995) and April 1979-1983 (Henwood, pers. comm.) were also obtained. Total carapace length (1979-1983) was converted to minimum carapace length using the formula  $SCL_{min} = (SCL_{total} \times 0.9774) - 0.2552$  provided by Henwood (1987a); data were also converted from English (in.) to S.I. units (cm). Standard carapace length (1992) was converted to minimum carapace length using a relationship established for 1,408 loggerheads collected by our studies between 2000 and 2008:  $SCL_{min} = (SCL_{standard} \times 0.9822) - 0.2557$ .

Blood samples for assessing turtle sex (College of Charleston, Grice Marine Biological Laboratory) and genetic origin (University of South Carolina, Biology Department) were collected in vacutainer tubes (10ml lithium heparin and 5ml heparin-free, respectively) and processed at sea for pending shore-based analyses.

Processing for sex determination involved centrifugation for five minutes, pipette extraction and transfer into 2.0ml cryovials, and storage in liquid nitrogen aboard ship until transfer to a shore-

based ultra-cold freezer. In the laboratory, plasma (50-500 $\mu$ l volume) was extracted using 4ml diethyl ether, and antibody (200 $\mu$ l) and titrated testosterone (~12000 cpm in 100 $\mu$ l) were added to duplicate samples and standards. Charcoal was added to all tubes, and centrifugation was used to separate bound and unbound fractions. Radioactivity was recorded with a liquid scintillation counter (Wallace 1409), and testosterone concentration measured using RIA MENU Software (P. Licht, University of California, Berkeley). Testosterone concentrations were corrected to account for extraction efficiency (85.2 to 97.1%) and the fraction aliquoted from the reconstituted sample (40%). Testosterone concentrations <200 pg/ml (2000-2003) or <350 pg/ml (2008) for loggerheads  $\leq 75.0$ cm SCLmin or  $\geq 85.1$ cm SCLmin were classified as female. Sex was not assigned for loggerheads between 75.1 and 85.0cm SCLmin due to high probability of error during this reproductive maturation window. In addition to testosterone, turtle size, proportionate tail length, and time of year were also used to determine sex for loggerheads  $\geq 85.1$ cm SCLmin given that nesting adult females also exhibit high testosterone values.

Processing for genetic determination involved syringe extraction and transfer of 0.5ml of whole blood into a 10ml plastic screw-top tube containing 5ml of lysis buffer solution; inverting and gently mixing five times; and room temperature storage. In the laboratory, a 380-base-pair (bp) fragment of mitochondrial DNA control region was sequenced and PCR amplified using primers CR-1/CR-2 and amplification conditions described in Norman et al. (1994). Haplotype assignments were matched to designations maintained by Archie Carr Center for Sea Turtle Research (ACCSTR) Genetics Bank.

#### *Data analysis*

Moran's Index (ArcGIS ArcInfo Desktop 9.2; ESRI, Redlands, CA) was used to statistically test for spatial differences in sampling and catch locations among years (2000-2003, 2008), as well as between fishery-dependent and fishery-independent sampling (2000-2003) between 32.650°N and 32.850 °N. Moran's Index measures feature similarity based on both the feature locations (x, y coordinates) and feature attribute values. With any set of feature locations, and a single attribute, the Index measures whether the location pattern is clustered, dispersed, or random based on that attribute. The Moran's Index value runs from +1.0 to -1.0. Values closest to +1.0 indicate clustering, and those closest to -1.0 indicate dispersion. The Z score produced is a measure of standard deviation, therefore very large or very small (negative) Z scores indicate that the values are in the tails of the distribution, making the pattern unlikely to be random.

Size distribution data for loggerheads in all studies were not normally distributed; thus, Kruskal-Wallis (Minitab 15®; Minitab, Inc.) or Chi-square (MS Excel, 2007) tests were used. In 2008, trawling was conducted at randomly selected location (May-July) as well as at targeted locations (August); size data for a given sub-region were not statistically different (Table 1.1) between randomized and targeted trawling, so data were pooled to increase sample size. Size distribution data for Kemp's ridley sea turtles was normally distributed; thus, one-way Analysis of Variance (ANOVA) was used (Minitab 15®) to test for statistical differences among years and studies.

Three designators (male, female, unknown) for sex data were possible; thus, data were not normally distributed and statistical testing was performed using Chi-square (MS Excel 2007). Due to the high probability of error in sex determination for loggerheads between 75.1 and

85.0cm SCLmin (the size bin during which maturity occurs; NMFS & USFWS, 2008), sex ratio for loggerheads in this size range was not analyzed.

Although more than 50 standardized haplotypes for Atlantic and Mediterranean loggerheads are maintained by the ACCSTR Genetics Bank, two haplotypes (CC-A01 and CC-A02) generally accounted for at least 85% of all haplotypes observed in our samples; thus, data were not normally distributed. Remaining haplotypes were consolidated as “other”, and genetics data were analyzed statistically using Chi-square analysis (MS Excel).

## **Results**

### *Species composition*

Trawling in the Charleston, SC, shipping entrance channel between 2004 and 2007 yielded 220 individual loggerheads, including five individuals captured twice during the same year and three individuals captured twice during successive years. Only two Kemp’s ridleys and one green sea turtle were collected from this location between 2004 and 2007.

Trawling in the Port Canaveral, FL, shipping entrance channel between 2006 and 2007 yielded 158 loggerhead collections (42 adult males, 9 adult females and 107 juveniles). Three adult males were recaptured during this study. Due to the study focus on adult males and high catch rates for both adult male and juvenile loggerheads, 72 of 107 juvenile loggerheads were not tagged before release; thus, it was unclear if any of these turtles were collected more than once. No other sea turtle species were collected from the Port Canaveral channel during 2006-2007.

Regional trawling in 2008 yielded 209 individual loggerheads, eight Kemp’s ridley sea turtles, and one green sea turtle. Loggerhead collections in 2008 included two turtles tagged by previous studies, two turtles captured twice during summer 2008, and the recapture of a turtle originally tagged and released during the regional survey in 2001.

### *Spatial distributions*

Inter-annual differences (2008 vs. 2000-2003) in sampling location were not detected for the regional survey (Table 1.2). However, locations for fishery-dependent (2000-2003) and fishery-independent sampling (2000-2003, 2008) sampling between 32.650°N and 32.850°N were determined to be spatially distinct ( $p < 0.001$ , Table 2). Significant clustering of loggerhead catch was detected among years off GA and SC (Table 1.2). Sampling off the coasts of SC (Figure 1.1a) and GA (Figure 1.1b) occurred over wider longitudinal extents than off FL (Figure 1.1c), where loggerhead clustering was not detected.

Loggerheads were collected throughout the regional trawl area in 2008; however, spatial disparities were evident. More loggerheads were collected between St. Augustine, FL, and Brunswick, GA ( $n=79$ ) than were collected throughout the extent of sampling off the SC coast ( $n=70$ ); however, 32% ( $n=25$ ) of loggerheads collected between St. Augustine, FL, and Brunswick, GA, in 2008 were collected during targeted sampling at “hot spots”. Twice as many loggerheads ( $n=60$ ) were collected between Brunswick, GA, and Savannah, GA, than were

caught in either sub-region off SC ( $n=30$  to  $40$ ); however, 20% ( $n=12$ ) of loggerheads collected between Brunswick, GA, and Savannah, GA, were collected during targeted sampling efforts. Kemp's ridley sea turtles were collected throughout the regional trawl area in 2008; however, half ( $n=4$ ) of the Kemp's collections occurred between St. Augustine, FL, and Brunswick, GA. Kemp's ridley collection locations in 2008 occurred in the general vicinity as 64 Kemp's ridley collections in our various studies since 2000 (Figure 1.2).

A single green sea turtle was collected off the SC coast in the general vicinity as the other two collections of this species off the coast of SC since 2000 (Figure 1.2). Only 10 green sea turtles have been collected by our various studies since 2000.

#### *Size distributions*

A significant (K-W,  $df=3$ ,  $p<0.001$ ) latitudinal gradient in loggerhead size distributions was noted throughout the regional area in 2008. Pair-wise comparisons revealed that median size of loggerheads collected between St. Augustine, FL, and Brunswick, GA (64.5cm SCLmin) was significantly smaller ( $p<0.001$ ) than median size of loggerheads collected between Brunswick, GA, and Savannah, GA (67.7cm SCLmin), Savannah, GA, to Charleston, SC (69.5cm SCLmin) and Charleston, SC, to Winyah Bay, SC (69.4cm SCLmin). Loggerhead size distribution between Brunswick, GA, and Savannah, GA, was only significantly different ( $p<0.001$ ) from loggerhead size distribution between Savannah, GA, and Charleston, SC.

Median size of loggerheads within the regional study area has generally increased since 2000 (Figure 1.3); however, increases in median size were only significant for loggerheads collected between Brunswick, GA, and Savannah, GA (K-W,  $df=4$ ,  $p<0.001$ ) and between Charleston, SC, and Winyah Bay, SC (K-W,  $df=4$ ,  $p=0.004$ ).

Eighty-five percent of loggerheads ( $n=219$ ) collected during fishery-independent sampling in the Charleston, SC, shipping entrance channel were between 55.1 and 75.0cm SCLmin. Size distribution was not significantly different ( $\text{Chi-sq}_{\alpha=0.05, df=15}$ ,  $X=1.62$ ,  $p=0.613$ ) among years.

Median size of loggerheads collected by fishery-dependent sampling ( $n=102$ , 63.0cm SCLmin) was significantly smaller (K-W,  $df=2$ ,  $p<0.001$ ) than median size of loggerheads from the Charleston, SC, shipping entrance channel ( $n=218$ , 66.6cm SCLmin) during 2004-2007 and loggerheads ( $n=199$ , 69.3cm SCLmin) collected by fishery-independent sampling near Charleston, SC (strata 43-48) during 2000-2003 and 2008.

Size of adult male loggerheads collected from the Port Canaveral, FL, shipping entrance channel was not statistically different between 2006 and 2007 (K-W,  $df=1$ ,  $p=0.779$ ); thus, data were pooled to characterize size frequency distribution in 5-cm increments (Figure 1.4). Adult male loggerhead size in April 2006-2007 was not statistically different (K-W,  $df=2$ ,  $p=0.189$ ) from April 1992 or April 1979-1983; however, only 20% of adult males collected in April 1979-1983 were  $\leq 90.0$ cm SCLmin compared to 39-48% of adult male loggerheads collected in 1992 and 2006-2007, respectively. Proportionately fewer adult male loggerheads  $\geq 100.1$ cm SCLmin were collected in 1979-1983 (10%) than in 2006-2007 (15%), with 10 (of 100) and 7 (of 42) adult male loggerheads  $\geq 100.1$ cm SCLmin collected in those year groupings, respectively.

Size distribution for eight Kemp's ridley sea turtles collected in 2008 (mean = 45.9cm SCLmin, range = 34.2 to 54.9cm) was not significantly different (ANOVA,  $df=4$ ,  $p=0.940$ ) than size distribution for 58 Kemp's ridley sea turtles (mean = 45.2cm SCLmin, range = 27.0 to 62.1cm) collected from the regional trawl survey area between 2000 and 2003. Two Kemp's ridley sea turtles (54.2cm SCLmin and 29.6cm SCLmin) collected from the Charleston, SC, shipping entrance channel also fell within the range of observed sizes for Kemp's during regional sampling and fishery-dependent sampling near Charleston, SC, ( $n=3$ ; 24.7 to 41.6cm SCLmin) and Brunswick, GA ( $n=10$ ; 27.5 to 56.1cm SCLmin).

Only two green sea turtles were collected between 2004 and 2008; a 28.6cm SCLmin individual was collected from the Charleston, SC, shipping entrance channel in August 2004 and 24.7cm SCLmin individual was collected during regional sampling off SC in July 2008. Size of green sea turtles collected in 2004 and 2008 was comparable (27.8 to 30.6cm SCLmin) to eight green sea turtles collected by fishery-dependent and -independent sampling between 2000 and 2003.

#### *Genetic distributions*

No significant difference was detected in the relative contributions of CC-A01, CC-A02 and "other" haplotypes among all loggerheads collected during the 2000-2003 regional survey, the 2008 regional survey, the 2004-2007 Charleston channel survey, the 2000-2003 Charleston observer study, and 2006-2007 trawling in the Canaveral channel (Table 1.3).

Within the regional survey, no significant differences in haplotype ratios were detected for years, states or sex (Table 1.4); however, a significant difference was noted for turtle size due to a greater observed frequency of occurrence for both CC-A01 and CC-A02 among loggerheads  $\geq 85.1$ cm SCLmin.

A significant difference among years was detected for the Charleston channel (Table 1.5). During 2004 and 2005, the ratio of CC-A01 to CC-A02 among loggerheads was 1.1 to 1 ( $n=162$ ), but increased to 3 to 1 in 2006-2007 ( $n=50$ ). No significant differences were noted in haplotype ratios among loggerheads collected from the Charleston channel with respect to collection month, turtle sex or turtle size. No significant differences were detected among haplotype ratios in the Canaveral channel with respect to turtle size or sex (Table 1.6). No significant differences in haplotype ratios were detected for the fishery-dependent survey among years; however, among juvenile male loggerheads, the ratio of CC-A01 to CC-A02 was significantly different and skewed towards CC-A01 at a ratio of 18 to 1 (Table 1.7).

Four haplotypes were observed among 29 Kemp's ridley genetic samples. LK1 accounted for 62% ( $n=18$ ) of all samples. LK3 accounted for 28% ( $n=8$ ) of all samples. LK2 ( $n=2$ ; 7%) and LK4 ( $n=1$ ; 3%) were infrequently observed, but both were present only in Kemp's ridley sea turtles  $<41$ cm SCLmin. No statistical difference in haplotype distributions for Kemp's ridleys were observed among years (Chi-square,  $df=3$ ,  $p=0.690$ ), sizes (Chi-square,  $df=3$ ,  $p=0.319$ ) or sex (Chi-square,  $df=3$ ,  $p=0.092$ ). Genetics samples were only collected for two green sea turtles, both of which were the CM1 haplotype.



### *Sex distributions*

Sex ratios for loggerheads  $\leq 75.0$  cm SCLmin were not significantly among sampling sub-studies (Table 1.8). Within the regional survey, significant differences among years were attributed to lower than expected occurrence of “unknown sex” turtles in 2001 and 2002 and higher than expected occurrence of “unknown sex” turtles in 2003 (Table 1.9).

Sex ratios for loggerheads  $\geq 85.1$  cm SCLmin were also not significantly different among the regional survey and the Charleston, SC, shipping entrance channel survey (Table 1.8); sex ratio was not assessed for loggerheads  $\geq 85.1$  cm SCLmin from the Port Canaveral, FL, channel survey given targeted collection of adult males, nor fishery-dependent surveys due to insufficient collection of large loggerheads. Within the regional survey, no significant differences were detected among years for loggerheads  $\geq 85.1$  cm SCLmin (Table 1.9).

No significant differences in sex by month or year were noted among loggerheads  $\leq 75.0$  cm SCLmin collected from the Charleston channel (Table 1.10). Insufficient sample size for loggerheads  $\geq 85.1$  cm SCLmin from this study location precluded their statistical consideration. Similarly, due to the small number of loggerheads  $\leq 75.0$  cm SCLmin that were processed in the Canaveral channel, and the skewed contribution of adult males from the same location, detailed examination of sex ratios for the Canaveral channel was foregone.

Sex for 72 Kemp’s ridley (55F: 10M: 7U) and seven green sea turtles (6F: 0M: 1U) was determined. No significant differences were noted for Kemp’s sex with respect to turtle size ( $<45$  cm vs.  $\geq 45.1$  cm; Chi-square,  $df=2$ ,  $p=0.430$ ). A significant difference in Kemp’s ridley sex ratio was noted among years (Chi-square,  $df=8$ ,  $p=0.029$ ), which was attributed to greater observed “unknown sex” Kemp’s in 2003 than predicted.

### **Discussion**

Larger turtles were more frequently collected in the northernmost sub-region and smaller turtles were more common in the south. The observed latitudinal size gradient contraindicates loggerhead stranding data from generally estuarine study areas between NY and the eastern Gulf of Mexico (GOM), from which Hopkins-Murphy et al. (2003) suggested developmental movement southward along the U.S. East Coast and into the eastern GOM. Given the importance of the Eastern Seaboard to multiple loggerhead stocks and natal homing behavior (Bowen et al., 2004), we propose that except for Long Island Sound, NY (Morreale et al., 1992; Burke et al., 1993), discrepancies in size distributions among geographically distinct study areas most likely reflects sampling location (and the timing of sampling). Smallest loggerheads in our studies tended to be collected closest to shore (i.e., northern FL) and in fishery-dependent sampling near the Charleston harbor entrance. Studies in estuarine water bodies (Epperly et al., 1995a, 2007; Erhardt et al., 2007) predominantly collect loggerheads slightly smaller (by ~5 cm) than what are typically encountered in our coastal surveys; however, direct comparisons of size distributions between adjacent estuarine and coastal habitats are lacking. Slightly larger mean sizes of loggerheads in Chesapeake Bay, VA (Lutcavage and Musick, 1985; Mansfield, 2006) may reflect the pseudo-coastal nature of this system and/or greater post-nesting use of Chesapeake Bay and its tributaries by adults. Similarly, larger median loggerhead size off SC likely reflects historically greater nesting activity than off GA (NMFS & USFWS, 2008).

Annual increase in loggerhead size was only statistically significant for selected portions of the regional trawl survey area, suggesting that growth of mid-sized, resident individuals is being tempered by changes in the frequency of occurrence of the smallest and largest individuals. In the absence of recruitment of small turtles, even the slowest reported loggerhead growth rates (Klinger and Musick, 1995; Parham and Zug, 1997; Bjørndal et al., 1998) should increase mean loggerhead size by at least 1-2cm annually, assuming that removal of the largest individuals does not occur concurrent with retardation of recruitment. Relatively stable median sizes throughout our regional survey area are contrary to observations of increasing mean loggerhead size in estuarine environments in NC (Epperly et al., 2007); however, several key distinctions should be made. First, data collection in NC occurred during different years (1995 to 2003 versus 2000 to 2008) and during a different season (i.e., fall) when loggerheads from VA to NY relocate to (Keinath, 1993; Morreale, 1999; Mansfield, 2006) and aggregate in and around NC sounds (Epperly et al., 1995a; Coles and Musick, 2000) during their fall migration. Second, size increases reported by Epperly et al. (2007) stemmed from a 5-cm dominant size class shift from 55 to 59cm SCLmin to a size class of 60 to 64cm SCLmin; however, even the 60-64cm SCLmin size class is smaller than the median size of loggerheads collected throughout our study area. A noticeable shift in predominance from 55-65cm SCLmin to 65-75cm SCLmin was noted for the Charleston, SC, shipping entrance channel from 1990-1993 to 2004-2007; thus, trends reported by this study for 2004-2007 and by Epperly et al. (2007) suggest that recruitment among smaller year classes less commonly collected in coastal surveys also remains strong.

Loggerhead size distributions observed in the Charleston, SC, shipping entrance channel during 2004-2007 were larger than distributions during 1990-1993. Only 39 of 219 (18%) of loggerheads collected from the Charleston, SC, channel in the current study were  $\leq 60$ cm SCLmin, compared to 30% (Van Dolah and Maier, 1993) and 44% (Dickerson et al., 1995) which were  $\leq 60$ cm SCLmin. Although sample sizes between earlier studies ( $n=45$  to 51 individual loggerheads) and the current study may also have contributed to different size frequency distributions between 1990-1993 and 2004-2007, similarity in size frequency distributions between the Van Dolah and Maier (1993) and Dickerson et al. (1995) studies suggests that despite small sample sizes, representative size frequency distributions were obtained during 1990-1993. Furthermore, although loggerheads  $\geq 75.1$ cm SCLmin in the current study were predominantly collected in May, loggerheads  $\geq 75.1$ cm SCLmin were collected between the spring and fall by Van Dolah and Maier (1993) and by Dickerson et al. (1995); thus, it is unlikely that the larger size distribution reported in the current study reflects sampling bias during periods when larger individuals may be more likely to be encountered. Therefore, given passage of more than a decade between studies, it would appear that strong age classes present in the early 1990's continue to persist, and that growth of turtles in these strong age classes has contributed to shifting size frequency distributions towards larger sizes.

Genetic ratios were similar for loggerheads collected in coastal waters from Port Canaveral, FL, to Winyah Bay, SC. Throughout the geographic range of sampling, genetic samples were dominated by the CC-A01 haplotype. Similarly, the ratio of CC-A01, CC-A02 and "other" haplotypes observed off the coasts of GA and SC were not different from ratios reported for 304 loggerheads stranded in these states during 1995-2001 (Bowen et al., 2004). Corroboration of haplotype frequencies reported for stranded turtles with data collected in this study for live, free-

swimming loggerheads from the same vicinity provides a critical stepping stone for extrapolating the results of Bowen et al. (2004). In addition to expressing that 86% of loggerheads collected from coastal waters between FL and the northeastern U.S. (NE USA) originated from the south FL (sFL) nesting colony, an equally important but less often disclosed observation by Bowen et al. (2004) is that throughout the range of loggerhead nesting in the Atlantic Basin, more hatchlings proportionately recruit (75% of offspring for the NEFL-NC nesting colony; 48-62% of offspring for seven other colonies) to the “northern foraging area” (FL to NE USA) than exclusively remain at foraging grounds associated with their respective nesting colonies. Thus, just as hatching success on sFL beaches drives the relative abundance of juvenile loggerheads in the “northern foraging area”, survival of loggerheads encountered in the northern foraging area has a disproportionate impact on future nesting of loggerheads throughout the Atlantic basin.

Subtle differences in haplotype ratios for loggerheads collected near Charleston, SC, may reflect important distributional shifts or might simply be an artifact of small samples sizes. Specifically, a pronounced change in the ratio of CC-A01 to CC-A02 was noted between 2004-2005 (when CC-A01 to CC-A02 was present at a 1.1 to 1 ratio) and 2006-2007 (when CC-A01 to CC-A02 was present at a 3:1 ratio). Similarly, juvenile male loggerheads collected during fishery-dependent trawling between 2000 and 2003 exhibited an 18:1 ratio between CC-A01 and CC-A02; however, only 19 total juvenile males were actually collected. If this ratio is indicative of natal homing for juvenile males on this foraging ground, our findings would seem counter to the assertion by Casale et al. (2002) of a male-biased dispersal for juvenile loggerheads in the pelagic phase between the Atlantic Ocean and the Mediterranean Sea, which was also based on a small sample size ( $n=65$  loggerheads). Assuming that the 18:1 ratio observed near Charleston, SC, is accurate, juvenile male loggerheads of U.S. origin may only briefly occupy habitats in the Mediterranean Sea given the small size of juvenile male loggerheads collected near Charleston, SC, and that all juvenile loggerheads of comparable size collected by bottom trawling ( $n=68$ ) in the Mediterranean Sea were reported to be of Mediterranean origin (Laurent et al., 1998).

Sex ratios among loggerheads were consistently biased towards females at a 2.5 to 1 ratio, which remained relatively stable across all size classes examined. Sex ratios reported here closely resemble sex ratios for juvenile loggerheads collected from estuarine and coastal waters between FL and NC (Wibbels et al., 1991; Schoop et al., 1998; Braun-McNeill et al., 2007). Sex ratios reported for neritic loggerheads in U.S. waters differs, however, from sex ratios determined using direct gonadal observation for (predominantly pelagic phase) loggerheads in the Mediterranean Sea, where a 1:1 ratio is reported (Casale et al., 2006). Braun-McNeill et al. (2007) provide a thorough discussion of the merits of monitoring sex ratios for species whose sex is determined during incubation and the implications of skewed sex ratios for effective management. One of the most pressing sex ratio issues pertains to discrepancy between hatchling sex ratios as high as 9:1 for hatchlings on FL beaches (Mrosovsky and Provancha, 1992) and the 2 to 3:1 ratio observed for pelagic and neritic phase loggerheads. Two schools of thought have emerged to explain these sex ratio discrepancies. The first proposes differential mortality of sFL female hatchlings, or females in general (Carthy et al., 2003; Hopkins-Murphy et al., 2003). Hopkins-Murphy et al. (2003) also suggest that there may be undiscovered female-biased foraging grounds in the poorly surveyed tropics, although they note that the northern foraging ground does seem to be the primary foraging area for neritic juveniles.

In addition to female-biased mortality, particularly stemming from near-lethal incubation temperatures (Carthy et al., 2003), male-biased dilution of the 9:1 ratio should also be included in the discussion of sex ratio discrepancies between hatchlings and other life history stages. Provided the underlying assumptions (Mrosovsky and Provancha, 1992; Bowen et al., 2004) are correct, 86 of every 100 loggerheads on the northern sampling ground should have originated from sFL and comprise 78 females and eight males. If all of the other 14 of 100 loggerheads which originated from beaches other than sFL were male, the resulting ratio would be 78 females to 22 males, or a 3.5:1 ratio. If sFL beaches contributed equally to the northern foraging ground genetic stock, other nesting colonies would need only to collectively produce males at a 3.6:1 ratio to achieve an overall 2.5:1 female to male ratio. However, because sFL beaches contribute six times (i.e., 86% vs. 14%) as many offspring to the northern foraging area, the collective male-bias from those other colonies would need to be 22 males for every female to achieve an overall 2.5:1 female to male ratio. Thus, rather than exclusively relying upon a strong male-bias to explain discrepancies we suggest that a combination of factors contributes to observing females 2.5x as frequently as males. First, by virtue of the fact that hatchlings enter the ocean off sFL with a 9:1 female-bias, predators lying in wait off of those beaches (Whelan and Wyneken, 2007) undoubtedly remove a considerable number of female hatchlings. Second, given limited observations of male-biased dispersal among juvenile loggerheads (Casale et al., 2002) and that 61% of sFL stock are reported to re-locate to the northern foraging area (Bowen et al., 2004), more than 10% of sFL turtles that shift to the northern foraging area may actually be male. Third, given limited evidence of male-biased sex ratios near Charleston, SC, in our study, the suggestion of habitat partitioning by sex is not entirely unreasonable. And lastly, if habitat partitioning by sex does occur, study area selection and sampling methodologies are also critical variables which must be taken into account.

Kemp's ridley and green sea turtles were also collected during our various research efforts since 2000, but represented just 5% and <1% of all sea turtles collected, respectively. Kemp's ridley sea turtles were collected throughout the spatial range of our sampling, but were collected most frequently between sGA and nFL. Distribution of Kemp's ridley sea turtles in our surveys is consistent with tag-recapture (Schmid, 1995) and satellite-telemetry (Gitschlag, 1996) data; however, Kemp's ridleys regularly occur as far north as Long Island Sound, NY (Morreale et al., 1992). In GA and nFL, green sea turtle collections occurred along the shore-ward boundary of our regional sampling area, and generally within 10km of major inlets. Using acoustic and radio telemetry, Renaud et al. (1995) tracked nine juvenile green turtles of similar size as collected in our studies and reported regular movements between coastal and estuarine environments via inlets, in their case a dredged shipping channel inlet. Off the SC coast, juvenile green sea turtles in our studies were only collected between Cape Romain and Winyah Bay, in the vicinity of where the sediment plume exiting Winyah Bay could be expected to occur. Trammel netting in estuarine habitats along the SC coast have documented only a handful of juvenile green sea turtles within Winyah Bay; however, 44% ( $n=65$  of 148) juvenile green sea turtles have been collected just to the south (Bulls Bay to Cape Romain) since 1992 (SCDNR, unpublished data 1). Juvenile green sea turtles are also the second most frequently occurring sea turtle species in estuarine collections in NC (Epperly et al., 1995a, 2007) and central FL (Erhardt et al., 2007).

**Table 1.1.** Summary of statistical testing (t-tests) for size differences between random trawling (May to July) and targeted trawling (August) events during summer 2008.

	<b>Florida</b>			<b>Georgia</b>			<b>South Carolina</b>	
	<u>Random</u>	<u>Recapture</u>		<u>Random</u>	<u>Recapture</u>		<u>Random</u>	<u>Recapture</u>
Mean SCLmin (cm)	65.4	65.6		68.8	69.5		71.9	75.6
Variance	40.4	49.8		42.4	50.5		54.7	69.8
Sample size	36	22		67	17		63	5
p-value, two-tailed	0.521			0.360			0.170	

**Table 1.2.** Summary of statistical testing (Moran's Index) for spatial clustering among sampling and turtle catch locations.

Area	Attribute	MI	Z score	P value	Pattern type
Regional	By years	-0.010963	-1.573739	0.116	
Regional	By catch	0.027705	4.139283	<b>3.5 x 10<sup>-5</sup></b>	Clustered
Regional	By total	0.041378	6.171967	<b>0.000</b>	Clustered
SC only	By years	0.002294	0.351236	0.725	
SC only	By catch	0.012184	1.520379	0.128	
SC only	By total	0.016533	2.042088	<b>0.041</b>	Clustered
GA only	By years	-0.041221	-1.926200	<b>0.054</b>	Dispersed
GA only	By catch	0.029655	1.442283	0.149	
GA only	By total	0.051585	2.489978	<b>0.013</b>	Clustered
FL only	By years	-0.038704	-0.857044	0.391	
FL only	By catch	-0.024338	-0.506860	0.612	
FL only	By total	-0.010076	-0.164247	0.870	
Charleston	By study	0.70305	15.50148	0.000	

**Table 1.3.** Chi-square test results for loggerhead haplotypes among study areas, 2000-2008.

Among studies

Chi stat	4.990		Haplotype	00 to '03	2008	Chs Chan	Chs Obs	Canaveral
df	8		CC-A01	404	105	112	51	39
Chi stat, df, p=0.05	15.507		CC-A02	304	69	80	28	26
p-value	0.759		Other	82	25	20	14	14

**Table 1.4.** Chi-square test results for loggerhead haplotype ratios within the regional trawl survey area, 2000-2003 and 2008.

Regional; inter-annual

Chi stat	9.822		Haplotype	2000	2001	2002	2003	2008
df	8		CC-A01	90	83	112	119	105
Chi stat, df, p=0.05	15.51		CC-A02	60	67	72	105	69
p-value	0.278		Other	21	24	19	18	25

Regional; turtle sex

Chi stat	8.015		Haplotype	F	M	U
df	4		CC-A01	305	167	32
Chi stat, df, p=0.05	9.488		CC-A02	254	92	25
p-value	0.091		Other	73	28	6

Regional; turtle size

Chi stat	10.475		Haplotype	<75	75to85	>85
df	4		CC-A01	425	44	38
Chi stat, df, p=0.05	9.488		CC-A02	326	32	12
p-value	<b>0.033*</b>		Other	97	6	3

**Table 1.5.** Chi-square test results for loggerhead haplotype ratios within the Charleston, SC, shipping entrance channel (2004-2007).

Charleston; inter-annual

Chi stat	13.233		Haplotype	2004	2005	2006	2007
df	6		CC-A01	54	22	27	9
Chi stat, df, p=0.05	12.592		CC-A02	48	20	9	3
p-value	<b>0.039*</b>		Other	16	2	2	0

Charleston; by month

Chi stat	1.246		Haplotype	May	Aug
df	2		CC-A01	69	20
Chi stat, df, p=0.05	5.991		CC-A02	48	10
p-value	0.536		Other	9	1

Charleston; turtle sex

Chi stat	5.730		Haplotype	F	M	U
df	4		CC-A01	81	28	3
Chi stat, df, p=0.05	9.488		CC-A02	66	10	4
p-value	0.220		Other	12	6	2

Charleston; turtle size

Chi stat	4.766		Haplotype	<75	75to85	>85
df	4		CC-A01	95	9	11
Chi stat, df, p=0.05	9.488		CC-A02	68	5	10
p-value	0.312		other	20	0	0

**Table 1.6.** Chi-square test results for loggerhead haplotype ratios within the Port Canaveral, FL, shipping entrance channel (2006-2007).

Canaveral; size groups				
Chi stat	1.498		Haplotype	≤75.0 cm    ≥75.1 cm
df	2		CC-A01	16    23
Chi stat, df, p=0.05	5.991		CC-A02	7    19
p-value	0.473		other	2    5

Canaveral; sex and ≥ 75.1 cm SCLmin				
Chi stat	5.408		Haplotype	M    F
df	2		CC-A01	20    3
Chi stat, df, p=0.05	5.991		CC-A02	15    4
p-value	0.067		other	2    3

Canaveral; sex and ≤ 75 cm SCLmin				
Chi stat	0.721		Haplotype	M    F
df	2		CC-A01	4    12
Chi stat, df, p=0.05	5.991		CC-A02	2    5
p-value	0.697		other	0    2

**Table 1.7.** Chi-square test results for loggerhead haplotype ratios within the fishery-dependent survey near Charleston, SC (2000-2003).

Charleston observer; inter-annual						
Chi stat	4.535		Haplotype	2000	2001	2002    2003
df	6		CC-A01	18	12	11    10
Chi stat, df, p=0.05	14.449		CC-A02	8	4	7    9
p-value	0.605		other	3	2	2    0

Charleston observer; by sex (all <75 cm)					
Chi stat	13.515		Haplotype	M	F    U
df	4		CC-A01	18	31    2
Chi stat, df, p=0.05	9.488		CC-A02	1	24    3
p-value	<b>0.009*</b>		other	0	6    1

**Table 1.8.** Chi-square test results for loggerhead sex ratios among studies, 2000-2008.

Among studies; ≤75.0 cm SCLmin							
Chi stat	14.085		Sex	2000-2003 R	2008 R	Chs Chan	Chs Obs    Canaveral
df	8		M	194	52	37	19    6
Chi stat, df, p=0.05	15.507		F	443	113	147	64    21
p-value	0.080		U	47	8	9	5    0

Among studies; ≥85.1 cm SCLmin							
Chi stat	0.127		Sex	2000-2003 R	2008 R	Chs Chan	Chs Obs    Canaveral
df	4		M	16	2	6	n/a    targeted
Chi stat, df, p=0.05	9.488		F	33	4	10	n/a    targeted
p-value	0.998						

**Table 1.9.** Chi-square test results for loggerhead sex ratios within the regional trawl survey; inter-annual variability.

Regional; inter-annual (<75)

Chi stat	41.358		Sex	2000	2001	2002	2003	2008
df	8		M	41	36	50	67	52
Chi stat, df, p=0.05	15.507		F	96	107	125	115	113
p-value	<b>p&lt;0.001</b>		U	12	1	4	30	8

Regional; inter-annual (<75 w/o unknown)

Chi stat	5.714		Sex	2000	2001	2002	2003	2008
df	4		M	41	36	50	67	52
Chi stat, df, p=0.05	9.488		F	96	107	125	115	113
p-value	0.222							

Regional; inter-annual (>85)

Chi stat	2.807		Sex	2000	2001	2002	2003	2008
df	4		M	3	6	2	5	2
Chi stat, df, p=0.05	9.488		F	10	10	8	5	4
p-value	0.591							

**Table 1.10.** Chi-square test results for loggerhead sex ratio within the Charleston, SC, shipping entrance channel survey (2004-2007).

Charleston channel; inter-annual (<75)

Chi stat	4.300		Sex	2004	2005	2006	2007
df	6		M	25	4	6	2
Chi stat, df, p=0.05	12.592		F	76	35	26	10
p-value	0.636		U	5	2	1	1

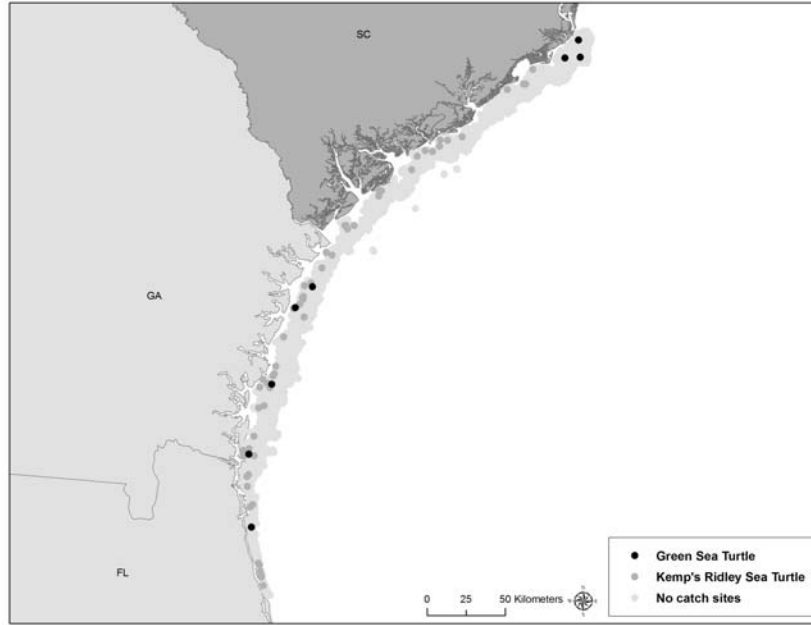
Charleston channel; by month (<75)

Chi stat	3.459		Sex	May	Jun	Aug
df	4		M	20	11	6
Chi stat, df, p=0.05	9.488		F	91	31	25
p-value	0.484		U	4	4	1

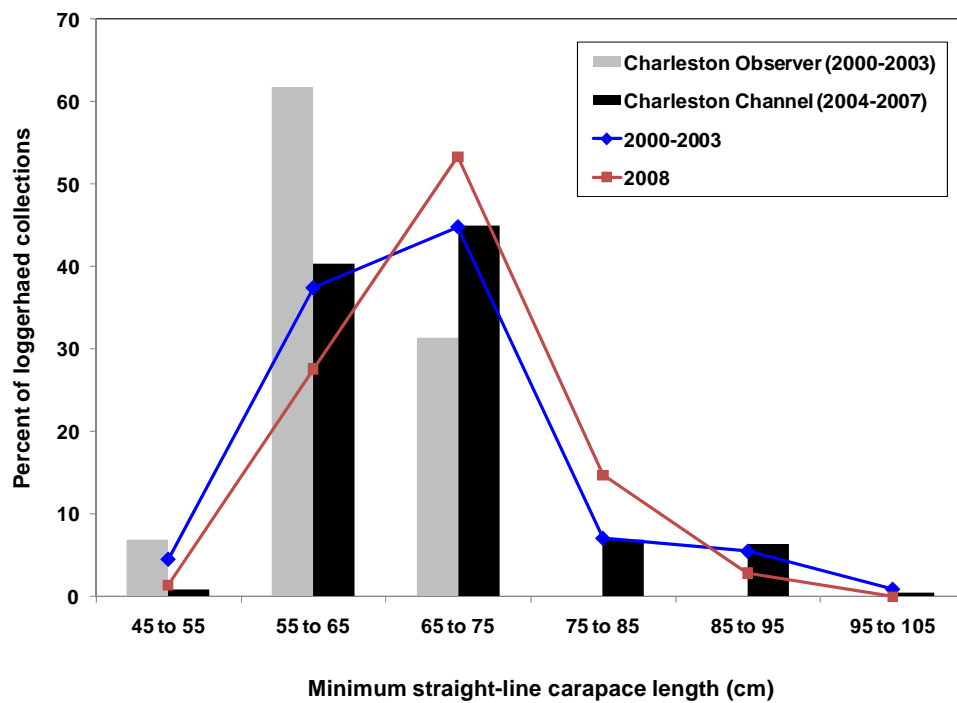




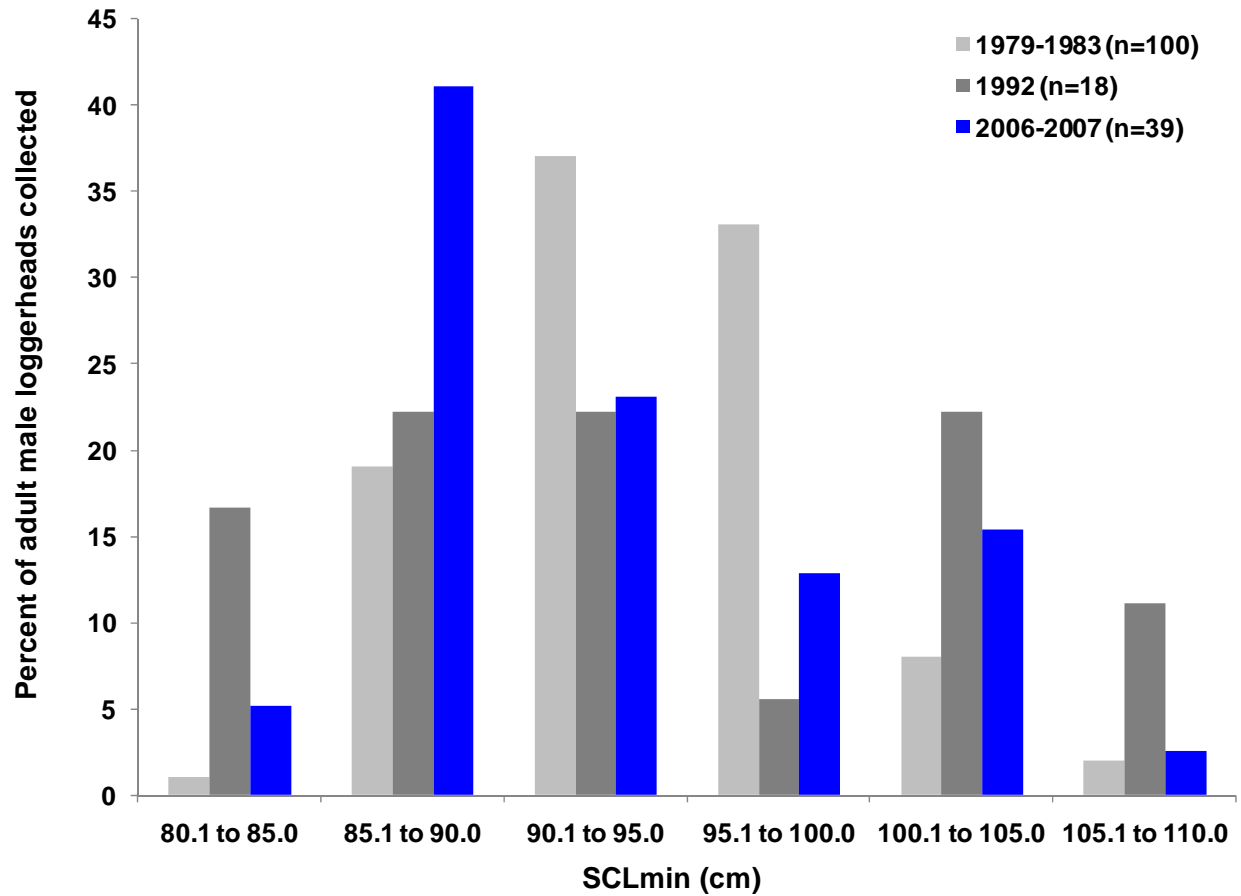
**Figure 1.1.** Spatial distribution of catch (black circles) and non-catch (gray circles) events for loggerheads off the coast of SC (A), GA (B) and northern FL (C); 2000-2003 and 2008.



**Figure 1.2.** Spatial distribution of green sea turtle catches (black circles), Kemp's ridley sea turtle catches (dark gray circles) and non-catch (light gray area) of either species during the regional trawl survey (2000-2003, 2008).



**Figure 1.3.** Size frequency distribution of loggerheads collected during the regional trawl survey (2000-2003, 2008), fishery-dependent sampling near Charleston, SC (2000-2003) and fishery-independent sampling from the Charleston, SC, shipping entrance channel (2004-2007).



**Figure 1.4.** Size frequency distribution of adult male loggerheads collected from the Port Canaveral, FL, shipping channel in April 2006-2007, with historical comparison to adult male loggerheads collected in April 1979-1983 (Henwood, 1987a) and 1992 (Dickerson et al., 1995).

## **Chapter 2 Sea turtle catch rates and factors affecting catch rates from randomized trawl surveys in coastal waters of South Carolina, Georgia, and northern Florida.**

### **Introduction**

For a variety of reasons, random sampling to collect sea turtles in coastal environments expends extensive effort relative to the number of turtles which are actually collected. First, life history stages occur throughout most habitat types across entire ocean basins (Carr, 1980); thus, populations are widely distributed. Second, non-catch events often exceed catch events for even seasonally abundant organisms from lower trophic levels such as plankton (Pennington, 1983); thus, as apex predators there should be relatively fewer sea turtles available to even be sampled. Similarly, the extraordinarily *k*-selected life history (Klinger and Musick, 1995) of sea turtles also limits their total abundance, and therefore opportunities for collection. Third, because they are adapted for movements on oceanic scales, their propensity for mobility may be manifested in a number of ways, including meandering along frontal boundaries (Polovina et al., 2001) and avoiding active collection gears such as trawls (Ogren et al., 1977). And lastly, especially prior to implementing a suite of protective measures since the 1970's, anthropogenic factors contributed (and in some cases still do) greatly to declines in abundance (Crouse et al., 1987).

Recognizing the importance of monitoring sea turtle populations in the water, the Second Revision to Recovery Plan for the Northwest Atlantic Population of the Loggerhead Sea Turtle (NMFS & USFWS, 2008) calls for establishing a network of long-term monitoring sites throughout the extent of foraging grounds. A regional trawl survey commissioned in 2000 (Maier et al., 2004) subsequent to similar recommendations made by the Turtle Expert Working Group and in-water workshop participants (TEWG, 1998; Bjorndal and Bolten, 2000) provides a critical baseline for such endeavors. Prior to the regional turtle survey, fishery-independent collections of sea turtles in coastal waters in the Southeastern USA (SE USA) primarily consisted of opportunistic observations during finfish surveys (Bullis and Drummond, 1978; SEAMAP-SA, 2000) or directed sea turtle trawling within shipping channels (Butler et al., 1987; Van Dolah and Maier, 1993; Dickerson et al., 1995), with greatest emphasis on the shipping channel for Port Canaveral, FL (Carr et al., 1980; Henwood, 1987b; Standora et al., 1993a,b; Dickerson et al., 1995). Fishery-dependent sea turtle data from the SE USA predominantly included fishery observer data from commercial shrimp trawling (Hillstead et al., 1977; Ulrich, 1978; Henwood and Stuntz, 1987; Schmid, 1995) and pound net fishing (Epperly et al., 2007).

The first objective of this chapter was to present spatial and temporal analyses of standardized catch-per-unit effort for loggerhead sea turtles collected during randomized trawling in the regional trawl survey area during 2008. Data collected from the same survey area in 2000-2003 (Maier et al., 2004) are included in statistical analyses, to provide the necessary context for evaluating data collected in 2008. The second objective of this chapter was to evaluate a suite of factors which may have potentially influenced catch rate data (in all years), including sampling design considerations and hydrographic and meteorological conditions at the time of sampling. Examination of factors which may have influenced catch rates expand upon similar analyses conducted by Maier et al. (2004) for the 2000-2003 data set.

## Methods

### *Data collection*

Trawling was conducted at randomly selected locations in coastal waters (4.6 to 12.2m deep) corresponding to the shoreline between Winyah Bay, SC, and St. Augustine, FL. Sampling was conducted between May and August 2000, and May through July in 2001-2003 and 2008.

Standardized fishing effort (i.e., 30.5m net-hour) was calculated for each sampling event (defined as port and starboard “turtle nets” only) as described by Jamir (1999):

Standardized effort = (# nets x net length/30.5) \* (event tow time in minutes/60)

Catch per unit effort (CPUE) was calculated for each sampling event as species-specific catch divided by standardized sampling effort for each sampling event. Permitted sampling event duration between 2000 and 2003 was 30 minutes; however, a change in our Federal Permit in 2008 reduced sampling event time to 20 minutes. As such, CPUE calculations in 2008 reflect a 33% (from 0.649 to 0.433 30.5m net-hr) reduction in fishing effort. Thirteen of 3,219 (0.4%) sampling events were excluded due to aberrant tow durations.

### *Station metadata and environmental data*

Standard station metadata consisted of vessel tow speed (kts) at the start of each trawl; two hydrographic parameters (surface water temperature (°C or converted to °C) and wave height (ft, converted to m); and three meteorological parameters (wind speed (kts), wind direction (text), and barometric pressure (millibars (mb) or converted to millibars). In 2000 and 2001, wave height was estimated as half the distance between trough and crest, but since 2002 has been estimated as the apparent distance between trough and crest. Because trawl transects were not always completely linear, calculating mean trawl tow speed as distance traveled divided by time trawled yielded spurious data; thus, initial tow speed at start of trawl was utilized. Surface water temperature was measured using a digital thermometer after obtaining a surface water sample at each station in 2001-2002; however, since 2003, surface water temperature has been recorded using a transducer located on each ship's hull approximately 1.5m below the water surface. Wind direction was converted to numeric as follows: N (0°); NNE (22.5°); NE (45°); ENE (67.5°); E (90.0°); ESE (112.5°); SE (135.0°); SSE (157.5°); S (180.0°); SSW (202.5°); SW (225.0°); WSW (247.5°); W (270.0°); WNW (292.5°); NW (315.0°); and NNW (337.5°).

Hourly observations of surface water temperature (SST, °C), wave height (m), barometric pressure (mb) and wind speed (kts) and direction (°) were obtained for three NOAA National Data Buoy Center (NDBC) data buoys (<http://www.ndbc.noaa.gov/maps/Southeast.shtml>). Off SC, data for the Edisto Buoy (#41004; 41nm (76km) SE of Charleston, SC; 34m water depth) was available for 2000-2003 and 2008. Off GA, data for the buoy at the Gray's Reef National Marine Sanctuary (GRNMS, #41008; 40nm (74km) SE of Savannah, GA; 18m water depth) was available for 2000-2003 and 2008. Off FL, data for the St. Augustine buoy (#41012; 40nm (74km) ENE of St. Augustine, FL, at 38m water depth) was only available for 2002, 2003 and 2008. Only buoy data collected on the day of sampling off the corresponding coast and between 1100 and 2300 UTC (0700 and 1800 local time) were utilized in analyses.

### *Data analyses*

Loggerheads were not collected in 77% of sampling events; thus, data could not be transformed to fit a normal distribution. CPUE calculated for each sampling event was tested using non-parametric statistics (Kruskal-Wallis and Dunn-Bonferroni pairwise comparisons, Minitab 15®, Minitab, Inc.). Maier et al. (2003) calculated CPUE on a vessel-day basis rather than a sampling event basis, which reduced the frequency of non-catch events from 77% to 21%; however, replicate observations were also reduced by 87% and the resulting data were still not normal.

CPUE was calculated for loggerheads and Kemp's ridleys. For loggerheads, CPUE was also calculated for five 10cm size classes (45.1 to 55cm SCLmin; 55.1 to 65cm SCLmin; 65.1 to 75cm SCLmin; 75.1 to 85cm SCLmin; and 85.1 to 95cm SCLmin). Only one loggerhead <45.1cm SCLmin and seven loggerheads >95.0cm SCLmin were collected during the regional survey; thus, although these turtles were included for overall CPUE calculations, CPUE for their respective size classes was not examined due to insufficient data.

CPUE was examined for the entire region as well as among four sub-regions. Sub-regions were selected based on strata codes used by the Southeastern Area Monitoring and Assessment Program (SEAMAP) as follows: St. Augustine, FL, to Brunswick, GA (strata 27-28 to 33-34); Brunswick, GA, to Savannah, GA (strata 35-36 to 39-40); Savannah, GA, to Charleston, SC (strata 41-42 to 45-46); and Charleston, SC, to Winyah Bay, SC (strata 47-48 to 49-50).

Principal Components Analysis (Minitab 15®, Minitab, Inc.) was used to characterize associations between loggerhead CPUE and 14 additional factors which may have affected catch rates. Three temporal factors included were year, Julian date, and time of day. Five spatial factors consisted of mean latitude and longitude of the trawl transect, mean water depth of the trawl transect, percent change in water depth during the trawl, and distance (km) from shore which was calculated using the "Near" analysis tool in ArcInfo 9.3 (ESRI). The final six factors included initial vessel towing speed (kts), wind speed (kts) and direction (numeric), barometric pressure (mb), water temperature (°C) and percent cloud cover.

## **Results**

Annual sampling effort ranged from 254.8 x 30.5m net-hours in 2008 (588 x 20 min stations) to 461.3 x 30.5m net-hours (714 x 30 min stations) in 2003 (Table 2.1). Mean annual sampling effort between 2000-2003 and 2008 was 389.9 x 30.5m net-hours. Nine hundred ninety loggerheads, 62 Kemp's ridleys and eight green sea turtles were collected.

### *Loggerhead CPUE*

Overall loggerhead CPUE was not significantly different (K-W, df=4, p=0.504) among years; however, significant differences in CPUE for some size classes were noted (Figure 2.1). CPUE for the 45.1 to 55.0cm SCLmin size class was significantly greater (K-W, df=4, p=0.004) in 2000 than in 2002, 2003 or 2008. CPUE for the 65.1 to 75.0cm and the 75.1 to 85.0 SCLmin size classes were significantly greater (K-W, df=4, p=0.002) in 2003 and 2008 than in 2000. CPUE for the 65.1 to 75.0cm SCLmin size class was also greater in 2003 than in 2001, and CPUE for the 75.1 to 85.0cm SCLmin size class was greater in 2008 than in 2001 or 2002.

Loggerhead CPUE was statistically different (K-W,  $df=2$ ,  $p<0.001$  to  $0.007$ ) among sub-regions overall as well as for most size classes (Figure 2.2). Overall and for loggerheads  $<65.1$ cm SCLmin, CPUE was greater between St. Augustine, FL, and Brunswick, GA, than for any other sub-region. CPUE for loggerheads  $65.1$  to  $75.0$ cm SCLmin was greater between St. Augustine, FL, and Brunswick, GA, than anywhere off the SC coast, as well as greater between Brunswick, GA, and Savannah, GA, than between Charleston, SC, and Winyah Bay, SC. CPUE for loggerheads  $75.1$  to  $85.0$ cm SCLmin was greater between Savannah, GA, and Charleston, SC, than for any of the other three sub-regions.

With one exception, inter-annual CPUE within sub-regions was not significant overall or for 10-cm size classes (Figure 2.3). Within the Brunswick, GA, to Savannah, GA, sub-region, CPUE for loggerheads  $65.1$  to  $75.0$ cm SCLmin was significantly greater (K-W,  $df=4$ ,  $p=0.001$ ) in 2003 and 2008 than in 2001, as well as greater in 2003 than in 2000. Lack of significant inter-annual differences within sub-regions is corroborated by similarity in intra-annual CPUE observations for each of the four geographic sub-regions as well (Figure 2.4).

#### *Kemp's ridley CPUE*

CPUE for Kemp's ridleys was not statistically different (K-W,  $df=4$ ,  $p=0.485$ ) among years; however, CPUE was statistically different (K-W,  $df=3$ ,  $p=0.023$ ) among sub-regions. Pairwise comparisons revealed Kemp's ridley CPUE was significantly greater between St. Augustine, FL, and Savannah, GA, than between Charleston, SC, and Winyah Bay, SC. Within sub-regions, significant inter-annual differences were not detected (K-W,  $df=4$ ,  $p=0.244$  to  $0.936$ ).

#### *In situ conditions during sampling*

Water temperature observations between data sets (vessel, oceanographic buoy) were generally in agreement, except for summer 2003 and summer 2008 when vessel-recorded temperatures were appreciably cooler than offshore buoys (SC sub-regions only, Figure 2.5). Coolest water temperatures were observed in summer 2003, and warmest water temperatures were observed in summer 2000 and 2001. Warmer vessel temperature records during 2000-2001 than observed in later years was partially attributed to different temperature measurement methodologies.

Discernible differences existed in barometric pressure readings between data sets, with vessel readings showing greater variability (within and among years) than buoy readings (Figure 2.6). Where both data sets were available, mean vessel readings were higher than buoy readings between St. Augustine, FL, and Savannah, GA. Vessel readings for barometric pressure for two sub-regions off the SC coast were generally comparable or lower than barometric pressure readings at the Edisto Buoy, except for in 2001 (Savannah, GA, to Charleston, SC, only).

Wind speeds recorded aboard vessels exhibited greater within and among year variability (with no consistent pattern) than buoy readings, which were generally stable among years (Figure 2.7). Wind direction was highly similar between vessel and offshore buoys, except for 2002 when SE winds were consistently recorded among vessels sampling south of Charleston, SC (Figure 2.8).

Wave height measured at all three oceanographic buoys exceeded wave height recorded aboard vessels (Figure 2.9). Wave height among two offshore buoys showed greater similarity among

years than with the GRNMS which was located closer to shore. Greatest wave heights aboard vessels were observed in 2002, and declined progressively between 2003 and 2008. Pronounced discrepancy between vessel wave heights in 2000 and 2001 is at least partially attributed to a correction of wave height estimation methods implemented in 2002.

Mean vessel speed ranged from 2.6 to 2.8kts among years and sub-regions (Figure 2.10). Lowest annual mean vessel speed for each of four sub-regions was observed in 2003. No other consistent patterns in vessel speed among years were noted.

#### *Principal Components Analysis*

Sixty-one percent ( $n=1950$  of 3206) of sampling events contained all hydrographic and meteorological parameters. Barometric pressure (45%,  $n=894$  records), water temperature (28%,  $n=532$  records) and wind direction (11%,  $n=214$  records) accounted for most absent data points. Among sub-regions, 52% of sampling events between Brunswick, GA, and Charleston, SC, were discounted for PCA because of missing environmental data; 32% of sampling events between Charleston, SC, and Winyah Bay, SC, and 18% of sampling events between St. Augustine, FL, and Brunswick, GA, were discounted for PCA because of missing environmental data. Significant differences (K-W,  $df=1$ ,  $p=0.001$  to  $0.047$ ) in CPUE were noted between retained sampling events and discounted sampling events between Brunswick, GA, and Charleston, SC; however, CPUE for retained events was slightly greater. Significant differences in CPUE for retained vs. discounted events were not detected elsewhere (K-W,  $df=1$ ,  $p=0.628$ - $0.643$ ).

Between St. Augustine, FL, and Brunswick, GA, principal components each accounted for between 18% (PC1) and <1% (PC15) of variance (Table 2.2). Strongest correlation with CPUE was an inverse relationship with sampling depth, with similar but slightly less pronounced inverse relationships with latitude, longitude and distance from shore (Figure 2.11). A moderately positive relationship was also noted between time of day (which was positively correlated with barometric pressure and inversely correlated with wind direction), as well as a moderately positive relationship with percent depth change during a sampling event.

Between Brunswick, GA, and Savannah, GA, principal components also each accounted for between 18% (PC1) and <1% (PC15) of variance (Table 2.2). Strongest correlations with CPUE were positive relationships with cloud cover and wind direction (auto-correlated) and water temperature (Figure 2.12). Strongest inverse relationship was between CPUE and Julian date. Station depth and distance from shore were auto-correlated with each other, but of little apparent relevance to loggerhead CPUE in this sub-region.

Between Savannah, GA, and Charleston, SC, principal components each accounted for between 20% (PC1) and <1% (PC15) of variance (Table 2.2). Strongest positive correlations with CPUE occurred with vessel speed, barometric pressure, Julian date and water temperature (Figure 2.13). Inverse relationships with CPUE were not observed; however, a strong inverse relationship between wind direction and cloud cover was observed. A strong positive relationship was noted between water depth and distance from shore and sampling year, as well as between station depth variation and time of day.



Between Charleston, SC, and Winyah Bay, SC, principal components each accounted for between 16% (PC1) and <1% (PC15) of variance (Table 2.2). A strong inverse relationship between CPUE and wind direction was observed, and which was less inversely related to cloud cover than was distance from shore (Figure 2.14). Cloud cover and time of day were the strongest positively correlated components with CPUE, followed by station depth variation. Station depth variation was inversely correlated with sampling year, and wind speed was inversely correlated with water temperature; however, with the exception of station depth variation, none of these components were strongly associated with CPUE.

## **Discussion**

Mean CPUE for loggerheads throughout the regional area in summer 2008 was 1.5 times greater than mean CPUE for loggerheads from the same area in summer 2000, when this randomized regional trawl survey was initiated. Despite a persistent inter-annual increase in loggerhead CPUE, significant differences in CPUE between 2000 and 2008 were not able to be detected for this relatively short-time series. However, significant differences in CPUE were noted with respect to some loggerhead size classes, which for the most part indicated significant increases rather than decreases (addressed elsewhere in this discussion). Similarly, based on standardized CPUE calculated by Maier et al. (2004) for historical data sets, loggerhead CPUE for the entire region in 2008 was 43 times greater than observed during coastal surveys in the South Atlantic Bight (SAB) between 1950 and 1976 (Bullis and Drummond, 1978), 14-17 times greater than observed in the shrimp fishery in FL and SC waters in the late 1970's (Ulrich, 1978; Henwood and Stuntz, 1987), and 23 times greater than fishery-independent surveys in coastal waters in the SAB in the late 1980's and early 1990's (Beatty et al., 1992). Thus, although increases in CPUE cannot automatically be assumed to represent increased abundance without first establishing the probability of loggerheads being present at the time of sampling (Anderson, 2001), it is highly improbable that CPUE increases of the magnitude documented for the SAB over the past 30 years could occur without an inherent and substantial increase in actual abundance.

Inability to detect trends in overall loggerhead CPUE between 2000 and 2008 was primarily attributed to large variance associated with annual CPUE values. In theory, as actual loggerhead abundance increases, so, too, would the variance associated with increasing CPUE values. For example, currently 77% of all trawling events yield no turtles; thus, variance is relatively small due to a single value (in this case zero) accounting for a large proportion of all CPUE values. As CPUE increases due to fewer non-turtle tows and/or greater frequency of multiple turtle tows, the variance associated with CPUE also inherently increases. Furthermore, increased variance associated with increased CPUE occurs independent of whether increased CPUE was attributed to actual change in abundance or just increased probability of collection. Lastly, due to trawl time restrictions of 20min bottom time in 2008 versus 30min bottom in 2000-2003, CPUE for all stations with turtles subsequently increased by 33% because a smaller denominator was used to calculate CPUE. Although shorter tow times may actually reduce the probability of turtle catch due to less time for a turtle to tire from trying to 'out swim the net' (Ogren et al., 1977) and therefore could be argued to represent conservative estimates of CPUE, an additional side-effect of shorter tow times is also an increase in variance for each successful loggerhead collection given that the CPUE would also be greater when the effort denominator is smaller.

Non-stable variance and non-normal distribution in our data set violated basic assumptions of linear regression, the basis of the TRENDS program (Gerrodette and Brandon, 2000). With those caveats in mind, Maier et al. (2004) used the 2000-2003 portion of this data set with TRENDS to determine that 16 years would be required to detect a 25% change in relative abundance. Epperly et al. (2007) effectively transformed their data to normal by treating zero catches separately (Pennington, 1983) and reported a 13.2% annual increase in CPUE over a nine year period. It is worth noting here that mean CPUE in the current study only increased by 47% between 2000 (0.442) and 2008 (0.652), a rate of change of only 6% annually and which was subsequently determined to be non-significant. Epperly et al. (2007) also estimated that 14 and 19 years were needed to detect a 10% annual increase for the less frequently collected green and Kemp's ridley sea turtles, respectively. Erhardt et al. (2007) reported a requirement of 24 years to detect a total change of 82% (3.4% annual rate of change) using TRENDS with data on juvenile loggerheads collected from the Indian River Lagoon in central FL; however, it was unclear if researchers resolved issues regarding non-normality of their data first.

In contrast to overall CPUE, significant annual increases in CPUE were detected for the most abundant size class (65.1 to 75.0cm SCLmin) and the size class (75.1 to 85.0cm SCLmin) during which maturation in loggerheads occurs (NMFS & USFWS, 2008). Inter-annual increase in mean CPUE for loggerheads 65.1 to 75.0cm SCLmin between Brunswick, GA, and Winyah Bay, SC, was progressive and sustained; however, inter-annual changes in mean CPUE for this size class were only significant between Brunswick, GA, and Savannah, GA. Between St. Augustine, FL, and Brunswick, GA, inter-annual increases in CPUE for loggerheads 65.1 to 75.0cm SCLmin occurred as a step function, with lowest mean CPUE in 2000, slightly greater and similar mean CPUE between 2001 and 2003, and a second slight increase in mean CPUE in 2008; however, increases were not statistically different. Similarly, inter-annual trends in mean CPUE for loggerheads 75.1 to 85.0cm SCLmin was progressive and generally sustained between Brunswick, GA, and Winyah Bay, SC, but resembled a step function between St. Augustine, FL, and Brunswick, GA. Furthermore, inter-annual increases in this size class were only significant across the region, and not within any particular sub-region. Thus, although the regional results are encouraging, it must be cautioned that the 75.1 to 85.0cm SCLmin size class only represents 8.5% ( $n=84$  of 990) of loggerheads collected in the regional survey, and rates of annual increases appear to be slower than for the 65.1 to 75.0cm SCLmin size class. Nonetheless, given natal homing of loggerheads (Bowen et al., 2004) and genetic and sex ratio (see Chapter 1) data which also suggest regionally localized loggerhead distribution, significant increases for this size class which represents mature and/or maturing loggerheads throughout the trawl survey area suggest that loggerhead nesting on SC and GA beaches are likely to improve within the next decade.

Less optimistically, CPUE for the 45.1 to 55.0cm SCLmin size class (3.6% of all loggerheads) decreased significantly between 2000 and 2008; however, decline in the smallest juveniles may not necessarily represent a decline in recruitment. First, juvenile loggerheads begin to transition from an oceanic to a neritic coastal existence at these sizes (Carr, 1980); thus, low frequency of occurrence of loggerheads in this size class should be expected. Second, the downward trend was only significant off GA, where just 17 loggerheads in this size class were collected over the five years of the regional survey. Fifteen of 17 (88%) of loggerheads collected in this size class off GA were collected during 2000 and 2001 (when 66% of loggerheads from this size class

were also collected throughout the entire study), and hydrographic conditions at the time of sampling may have increased the relative abundance of these smallest loggerheads above 'normal' levels. Specifically, Gulf Stream intrusion (Blanton et al., 1981) across the continental shelf of the South Atlantic Bight (SAB) was pronounced in 2000, concurrent with warmest SST and large mats of *Sargassum* and "tar balls" washing ashore (Arendt, personal observation). Thus, we propose that altered circulation patterns and/or changes in the distribution of food availability associated with hydrographic conditions at the time of sampling in 2000 (and to a lesser extent 2001) may have concentrated small sea turtles closer to shore. Conditions at the time of sampling may also have shifted smaller juveniles already located closer to shore further offshore, bringing them into the sampling area of the regional trawl survey. Given frequent collection of loggerheads in the 55.1 to 65.0cm SCLmin size class from the Charleston, SC, shipping channel (see Chapter 3) and significant increases in CPUE for similar-sized loggerheads in NC (Epperly et al., 2007), we suspect that recruitment (to locations not sampled by us) of loggerheads  $\leq 55.0$ cm SCLmin continues to remain strong. Prudence certainly dictates that researchers more likely to encounter loggerheads  $\leq 55.0$ cm SCLmin should continue to closely monitor trends in the relative abundance of these small loggerheads, however.

In addition to Gulf Stream influence in 2000, hydrographic conditions at the time of sampling in 2002 and 2003 may also have influenced catch rates off both SC and GA. In 2002, wind direction off both states shifted noticeably from S/SSW to SE, which was also associated with increased wave height (research vessel data only) and decreased water temperature, suggesting stormier conditions during sampling. Maier et al. (2004) recommended that sampling be suspended at wind speeds above 15 kts, due to significantly reduced catches of loggerheads attributed to less efficient gear operation and/or possible relocation of loggerheads to deeper and presumably calmer waters. Highest catch rates for loggerheads during fishery-dependent sampling near Charleston, SC, occurred in 2002 (Maier et al., 2004), and closer examination of those data reveals greater catch rates from within rather than out of the shipping entrance channel in both 2002 and 2003. Thus, natural and maintained channels may provide a deeper water refuge during storm events without the energy expenditure associated with relocating offshore. In 2003, a noticeable shift in wind direction from S/SSW to SW occurred off SC and GA, which was also associated with the lowest water temperatures noted aboard research vessels off both coasts. Maier et al. (2004) describe a coast-wide upwelling phenomenon which occurred in August 2003 as a result of increased freshwater runoff that began in the spring and was followed by a shift in the predominant wind direction. Indeed, the additional hydrographic data provided here support the assertion that pre-cursor conditions to this coastal upwelling event in 2003 may have shifted near shore loggerheads further offshore (and/or squeezed offshore loggerheads further inshore), directly contributing to increased capture (and re-capture) rates in 2003.

Results from principal components analysis were not consistent among sub-regions, and none of the components individually accounted for more than 20% of the total variance. Lack of complete data sets and subsequent removal of 17-50% of CPUE records may have partially contributed to inconclusive PCA findings; however, inconclusive findings also suggest that rigorous efforts will be needed to develop a reliable set of 'correction factors' for assessing the probability of spatial and temporal occurrence of loggerheads during sampling. Cloud cover was inversely related to wind direction and positively related to CPUE for several sub-regions;

however, cloud cover was not statistically significant across the entire region during 2000-2003 (Maier et al., 2004). Water depth and distance from shore were most strongly correlated with CPUE in the southernmost portion of the survey area; given that CPUE in this region was significantly greater than all other regions, the relative importance of water depth at the southernmost latitudes may have greatly contributed to the findings of significance for these parameters across the region for the 2000-2003 portion of the data set (Maier et al., 2004). Consequently, attempts to assess the probability of loggerhead occurrence may benefit the most from assessing regional and 'big picture' hydrographic influences, particularly in the context of density gradients (Epperly et al., 1995b) and subsequent effects on localized circulation patterns.

Kemp's ridley sea turtles were collected in just two percent of all sampling events. Catch rates were greatest in the southernmost sub-region; however, as noted by Maier et al. (2004), our sampling area probably does not equally survey optimal Kemp's ridley habitat. Sampling in the southernmost sub-region occurs closest to shore and most Kemp's ridleys throughout the region were collected on the inshore edge of our sampling area (Chapter 1). Ten Kemp's ridleys were caught by fishery-dependent sampling in near-shore waters in June 2000, with seven collected just offshore of St. Andrews Sound, GA, and three (of four collected during 2000-2003) collected near Charleston, SC. Although net design differed between fishery-dependent (4x40' nets; small mesh) and -independent (2x65' nets; large mesh) sampling, spatial discrepancies in sampling area between these two sampling designs further support the assertion of spatial bias against collection of Kemp's ridleys (Maier et al., 2004). Temporally, catch rates (Kemp's ridleys per 30.5m net-hr) in 2008 (0.028 +/- 0.3 SD) were comparable to 2000-2002 (0.025-0.032 +/- 0.2); however, higher catch rates (0.041 +/- 0.3) in 2003 likely stemmed from the same hydrographic conditions that we suspect also increased loggerhead CPUE.

The merits of this fishery-independent and directed turtle trawl survey in the southern portion of potentially the most important foraging ground for juvenile loggerheads in the NW Atlantic Basin (Bowen et al., 2004) cannot be over-stated. Considerable thought and experience melded a quality sampling design which meets most of the critical assumptions for assessing trends in abundance with low probability of producing hyper-stable or hyper-depleted assessments (Hilborn and Walters, 1992; Allen, 2000); however, additional assessments are needed to address detection probability (Anderson, 2001). To a large extent, the inability to detect a trend in CPUE for the overall data set appears to be a limitation of study duration and/or simultaneous increases between mean CPUE and associated variance. In the short-term, increases in CPUE for sizes representing mature and/or next in line to become mature is encouraging. Furthermore, the data presented herein provide a critical assessment tool which directly pertains to Recovery Objective #2 (in-water juvenile abundance increases at a faster rate than strandings for similar-sized turtles) and Demographic Recovery Criteria #2 (abundance increasing for at least one generation throughout a network of monitoring sites within the foraging ground) of the Recovery Plan for loggerheads in the NW Atlantic Turtle (NMFS & USFWS, 2008). Therefore, given the historic and recent successes of this study, as well as established mandates and definitions for recovery, we sincerely hope that this and parallel studies are permitted to flourish.

**Table 2.1.** Summary of trawling effort and loggerheads collected, 2000-2003 and 2008.

Year	N events	Effort (30.5m net hr)	N <i>Caretta</i>	Gross CPUE
2000	620	401.8	178	0.44
2001	602	389.7	181	0.46
2002	682	442.2	210	0.47
2003	714	461.3	255	0.55
2008	588	254.3	166	0.65

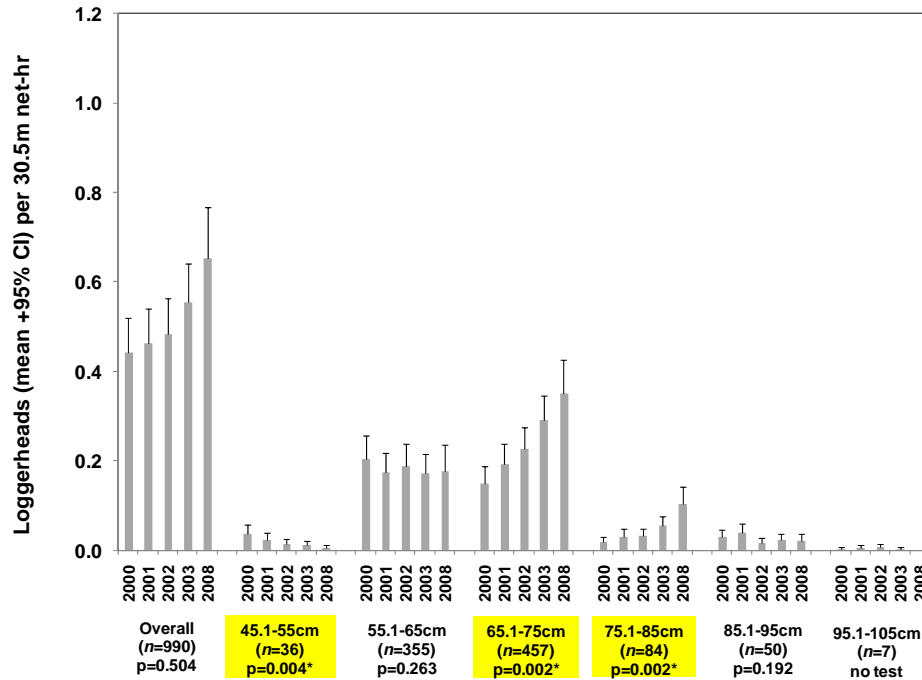
**Table 2.2.** Frequency distribution of variance accounted for by individual components determined by PCA for randomized trawling (2000-2003, 2008) between St. Augustine, FL, and Brunswick, GA (A); Brunswick, GA, and Savannah, GA (B); Savannah, GA, and Charleston, SC (C); and Charleston, SC, and Winyah Bay, SC (D).

"A"	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigenvalue	2.70	1.87	1.52	1.33	1.15	1.05	0.94	0.90
Proportion	0.18	0.12	0.10	0.09	0.08	0.07	0.06	0.06
Cumulative	0.18	0.30	0.41	0.49	0.57	0.64	0.70	0.76
	PC9	PC10	PC11	PC12	PC13	PC14	PC15	
Eigenvalue	0.82	0.71	0.60	0.57	0.46	0.34	0.04	
Proportion	0.06	0.05	0.04	0.04	0.03	0.02	0.00	
Cumulative	0.82	0.87	0.91	0.94	0.97	1.00	1.00	

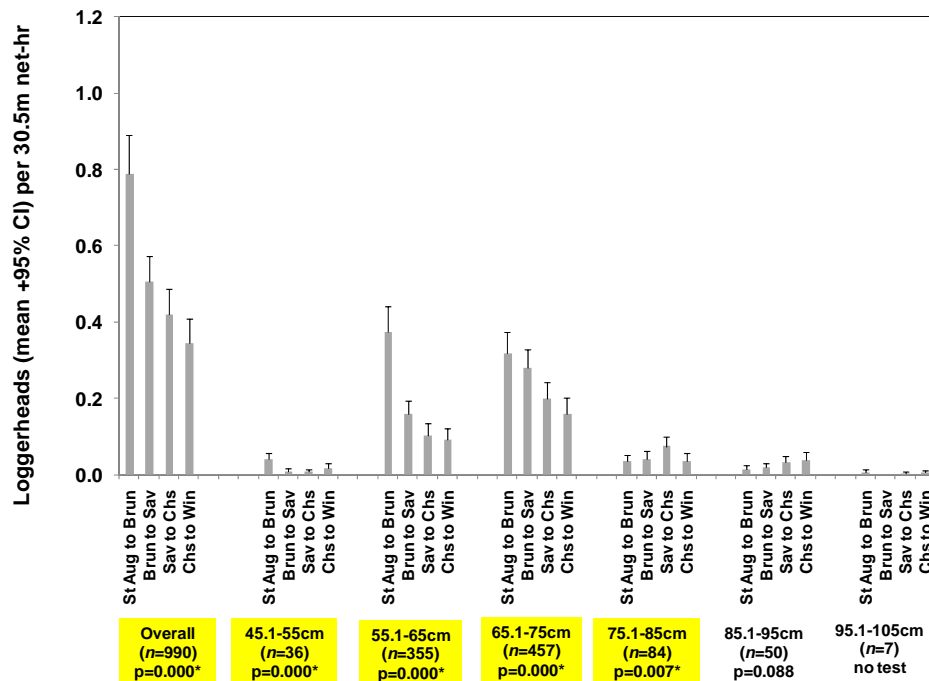
"B"	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigenvalue	2.63	2.07	1.73	1.41	1.15	1.05	0.97	0.84
Proportion	0.18	0.14	0.12	0.09	0.08	0.07	0.07	0.06
Cumulative	0.18	0.31	0.43	0.52	0.60	0.67	0.73	0.79
	PC9	PC10	PC11	PC12	PC13	PC14	PC15	
Eigenvalue	0.81	0.67	0.56	0.48	0.39	0.23	0.02	
Proportion	0.05	0.04	0.04	0.03	0.03	0.02	0.00	
Cumulative	0.84	0.89	0.93	0.96	0.98	1.00	1.00	

"C"	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigenvalue	3.01	2.03	1.64	1.28	1.21	1.06	1.02	0.95
Proportion	0.20	0.14	0.11	0.09	0.08	0.07	0.07	0.06
Cumulative	0.20	0.34	0.45	0.53	0.61	0.68	0.75	0.81
	PC9	PC10	PC11	PC12	PC13	PC14	PC15	
Eigenvalue	0.86	0.70	0.55	0.33	0.19	0.17	0.02	
Proportion	0.06	0.05	0.04	0.02	0.01	0.01	0.00	
Cumulative	0.87	0.92	0.95	0.98	0.99	1.00	1.00	

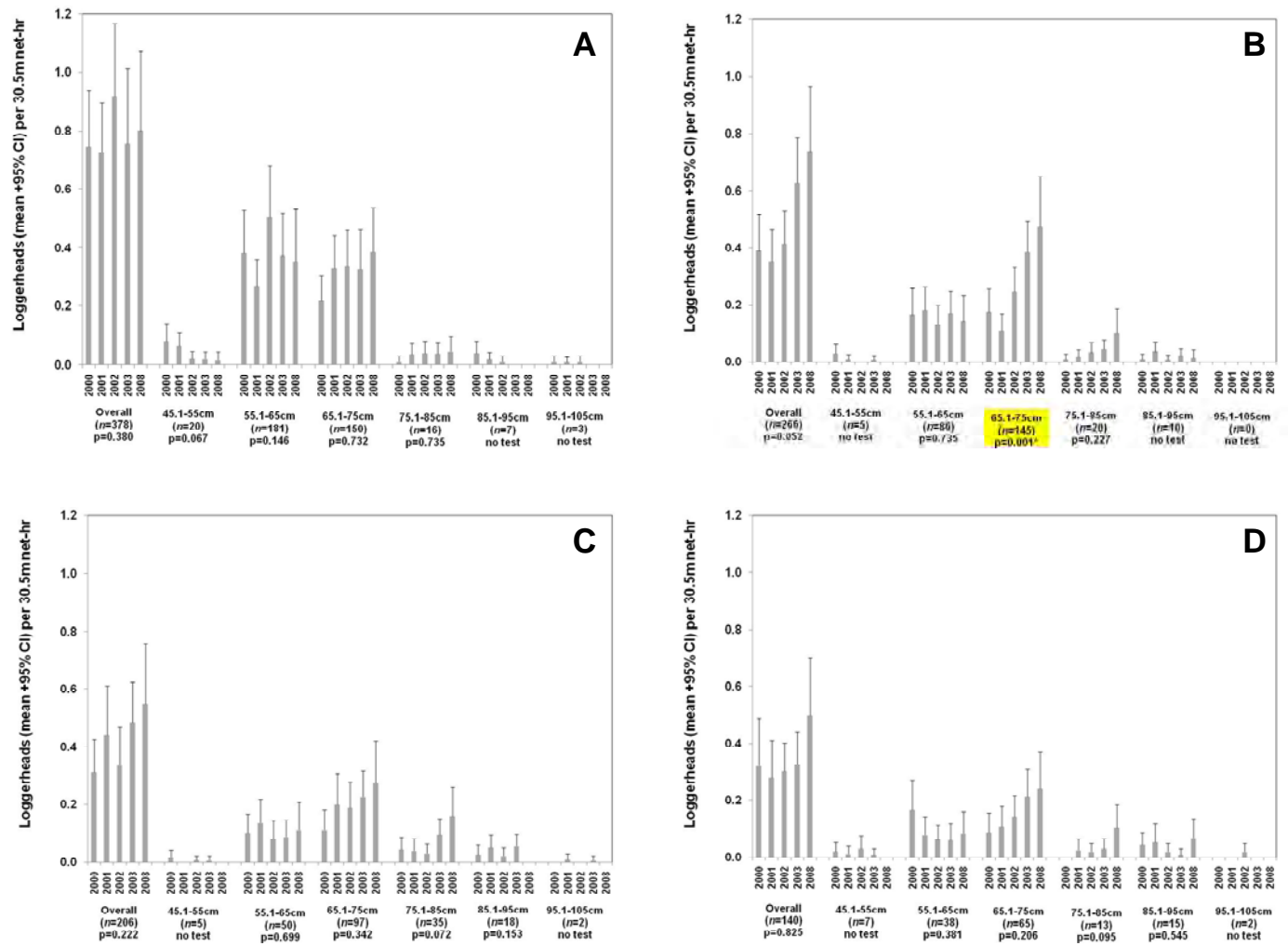
"D"	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigenvalue	2.42	2.30	1.74	1.47	1.30	1.12	0.99	0.85
Proportion	0.16	0.15	0.12	0.10	0.09	0.08	0.07	0.06
Cumulative	0.16	0.31	0.43	0.53	0.62	0.69	0.76	0.81
	PC9	PC10	PC11	PC12	PC13	PC14	PC15	
Eigenvalue	0.74	0.65	0.51	0.46	0.27	0.16	0.01	
Proportion	0.05	0.04	0.03	0.03	0.02	0.01	0.00	
Cumulative	0.86	0.91	0.94	0.97	0.99	1.00	1.00	



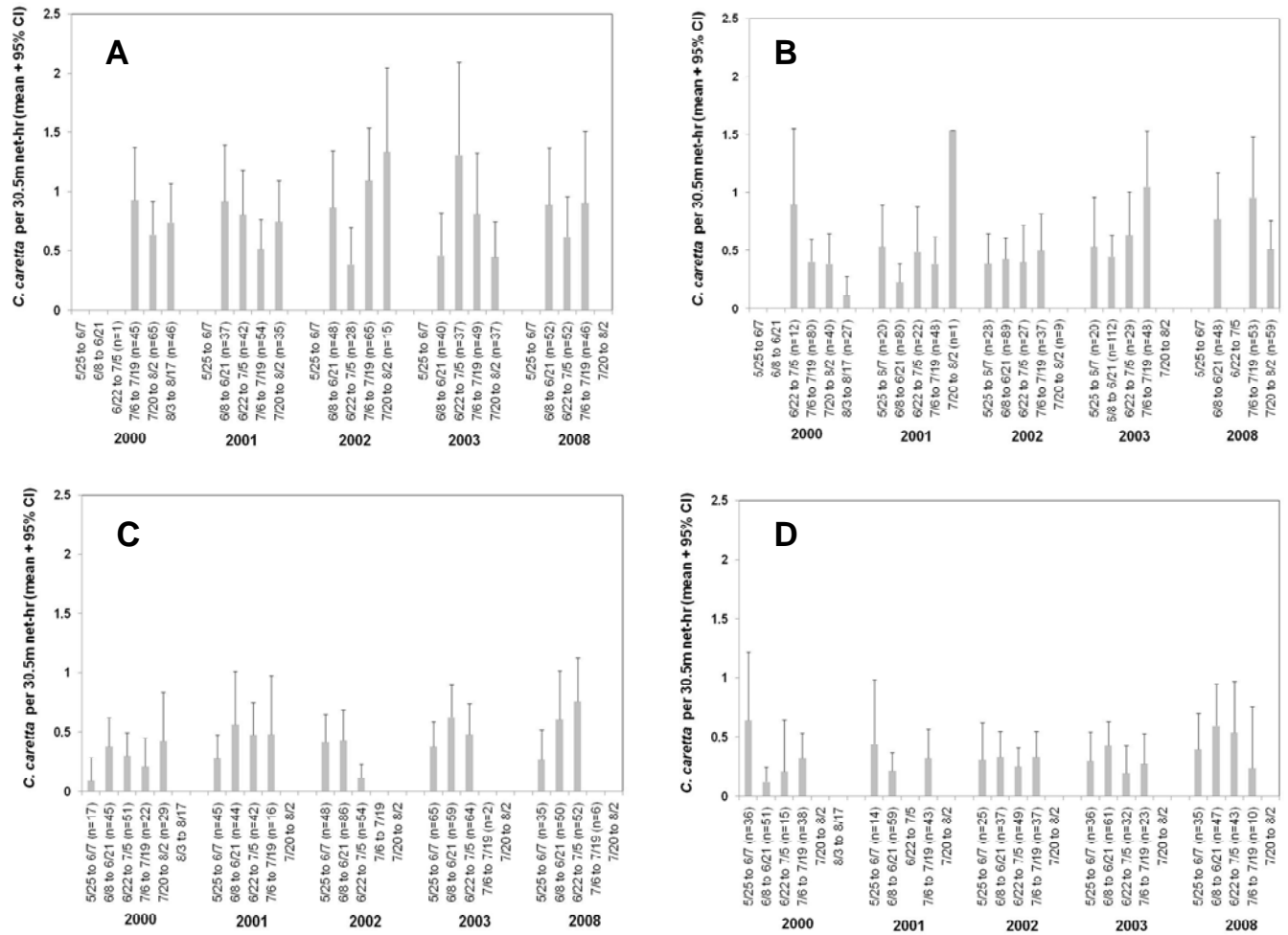
**Figure 2.1.** Temporal trends in loggerhead CPUE (overall and by 10-cm SCLmin size classes) throughout the regional turtle trawl survey area (2000-2003, 2008). Significance levels provided reflect Kruskal-Wallis rank testing; shaded groups with asterisk (\*) denote significant results.



**Figure 2.2.** Spatial trends in loggerhead CPUE (overall and by 10-cm SCLmin size classes) throughout the regional turtle trawl survey area (2000-2003, 2008). Significance levels provided reflect Kruskal-Wallis rank testing; shaded groups with asterisk (\*) denote significant results.

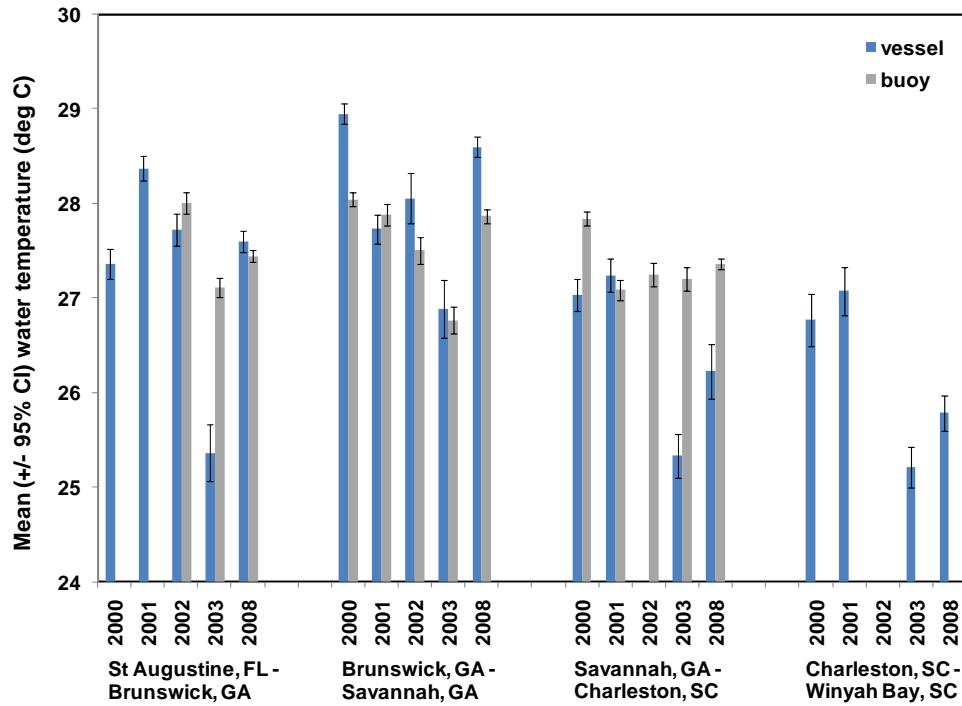


**Figure 2.3.** Temporal (2000-2003, 2008) trends in loggerhead CPUE (overall and by 10-cm SCLmin size classes) between St. Augustine, FL, and Brunswick, GA (A); Brunswick to Savannah, GA (B); Savannah, GA, to Charleston, SC (C); and Charleston, SC, to Winyah Bay, SC (C). Significance levels reflect Kruskal-Wallis testing; shaded groups with asterisk (\*) denote significant results.

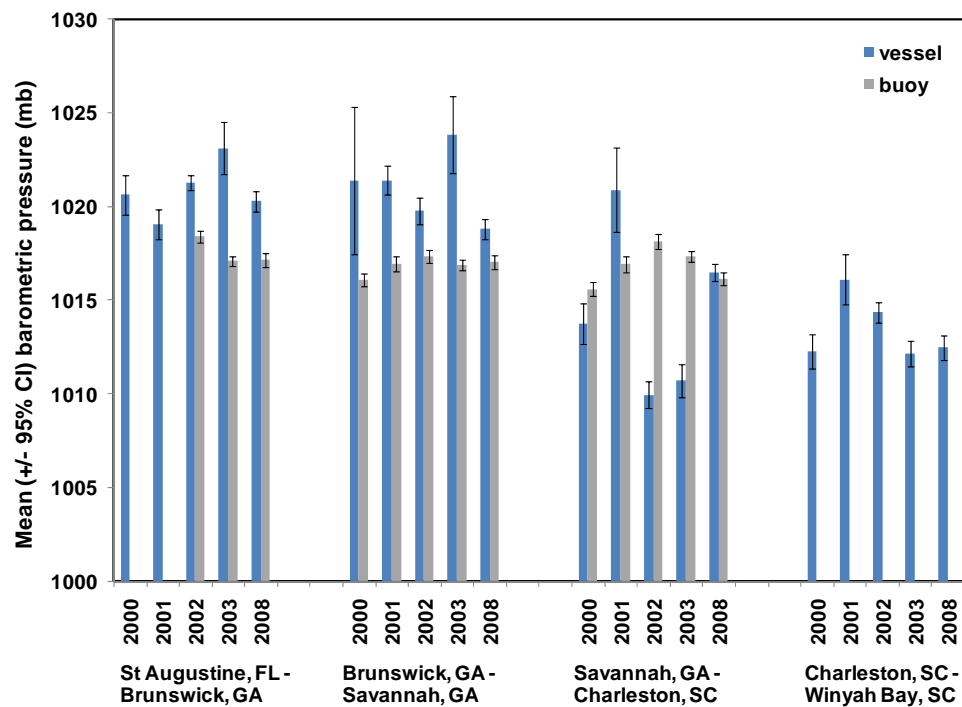


**Figure 2.4.** Intra- and inter-annual (2000-2003, 2008) trends in overall loggerhead CPUE between St. Augustine, FL, and Brunswick, GA (A); Brunswick to Savannah, GA (B); Savannah, GA, to Charleston, SC (C); and Charleston, SC, to Winyah Bay, SC (C).

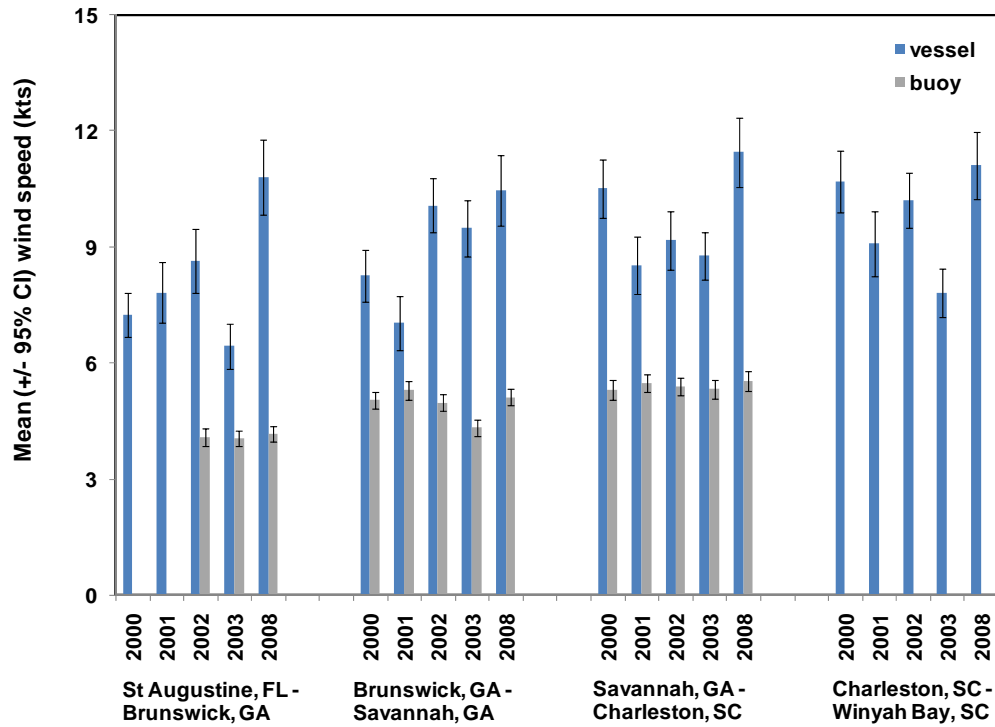




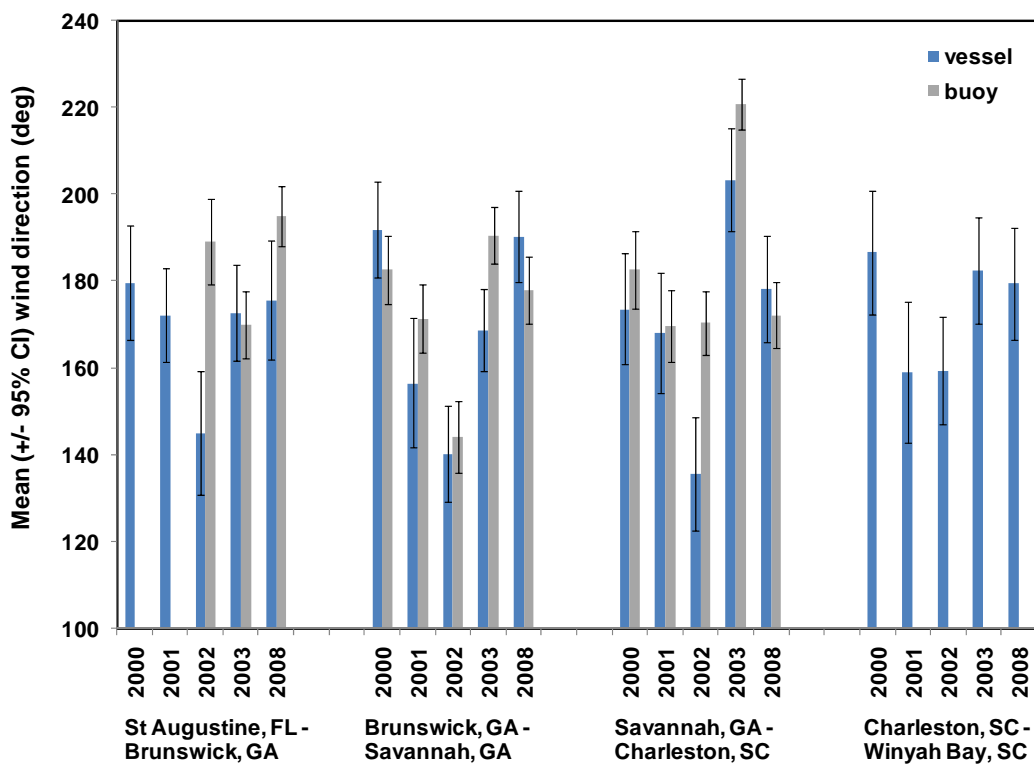
**Figure 2.5.** Mean ( $\pm$  95% C.I.) sea surface temperature during the regional trawl survey.



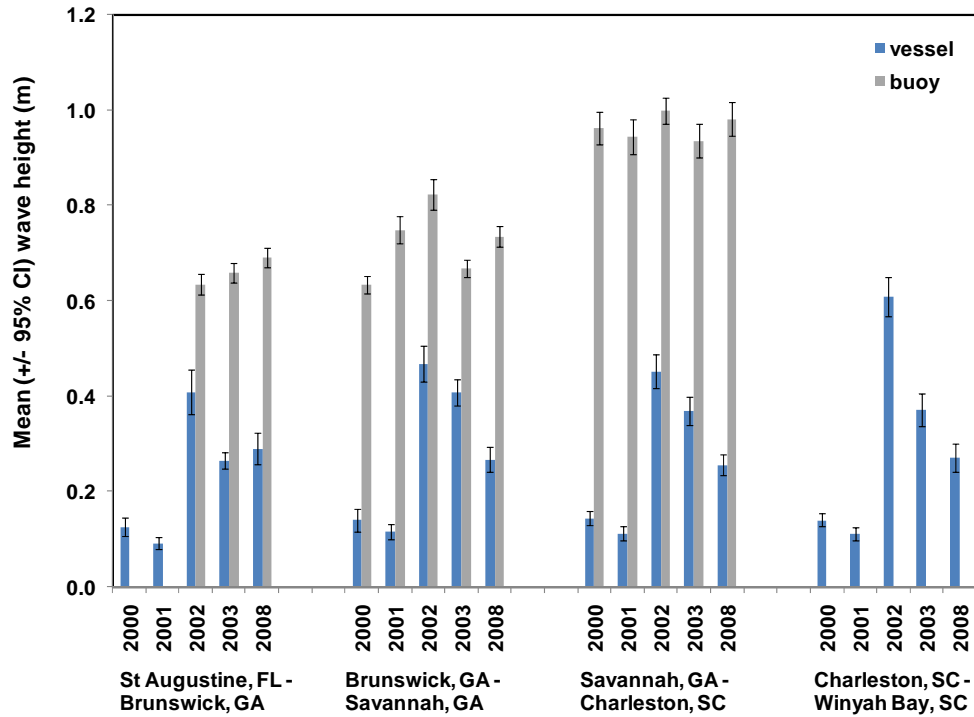
**Figure 2.6.** Mean ( $\pm$  95% C.I.) barometric pressure during the regional trawl survey.



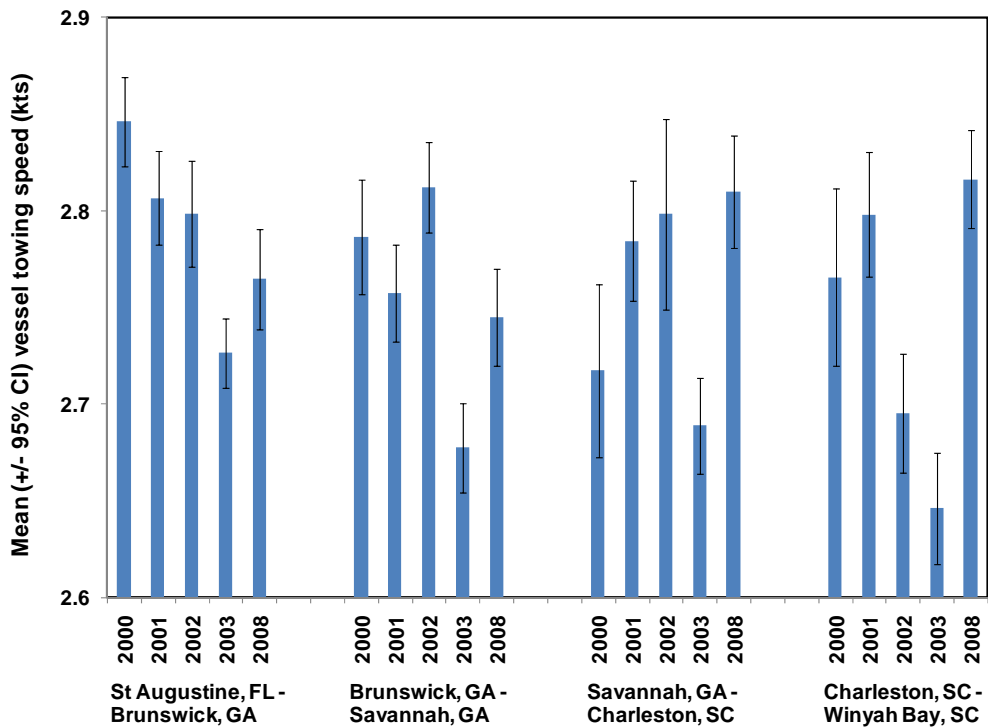
**Figure 2.7.** Mean ( $\pm$  95% C.I.) wind speed during the regional trawl survey.



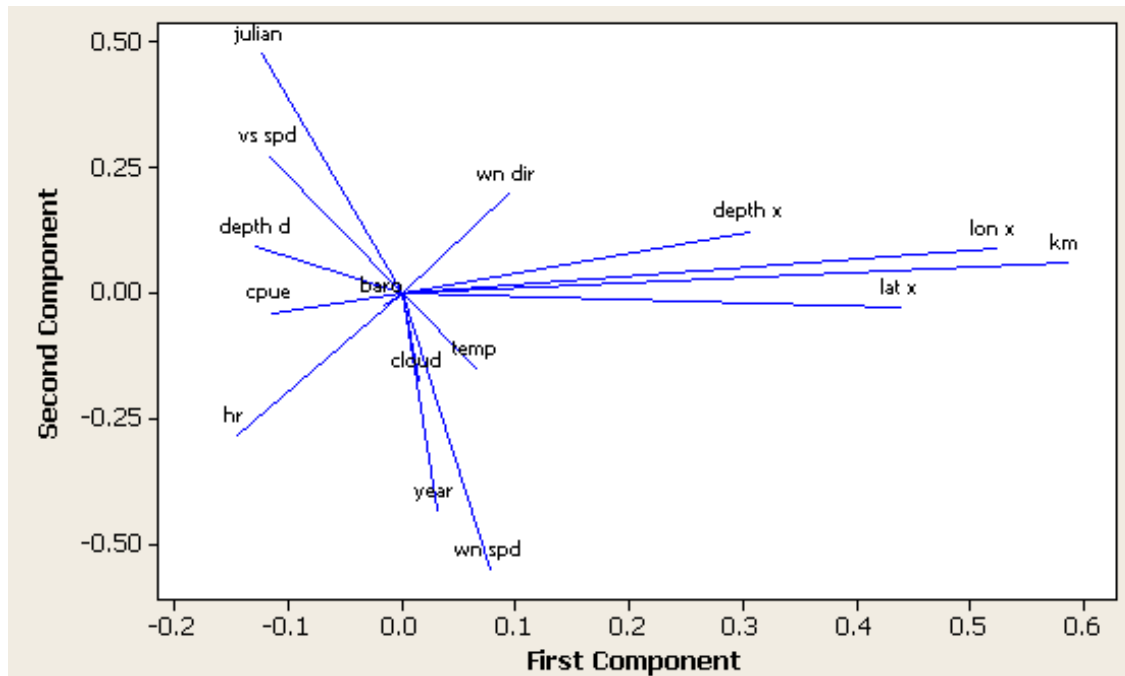
**Figure 2.8.** Mean ( $\pm$  95% C.I.) wind direction during the regional trawl survey.



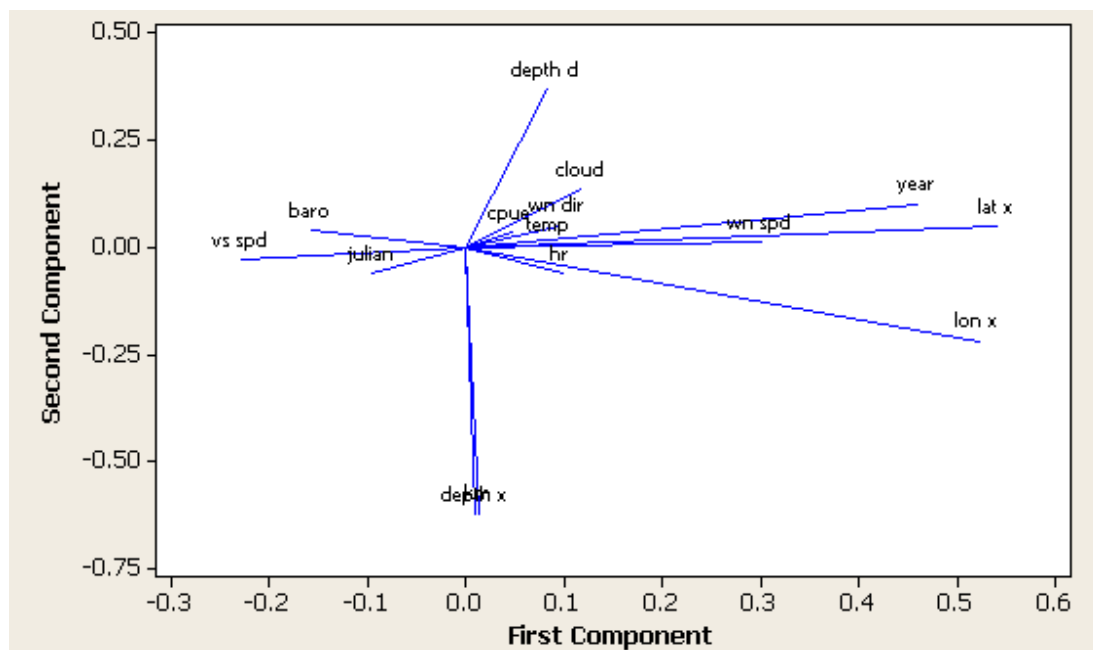
**Figure 2.9.** Mean ( $\pm$  95% C.I.) wave height during the regional trawl survey.



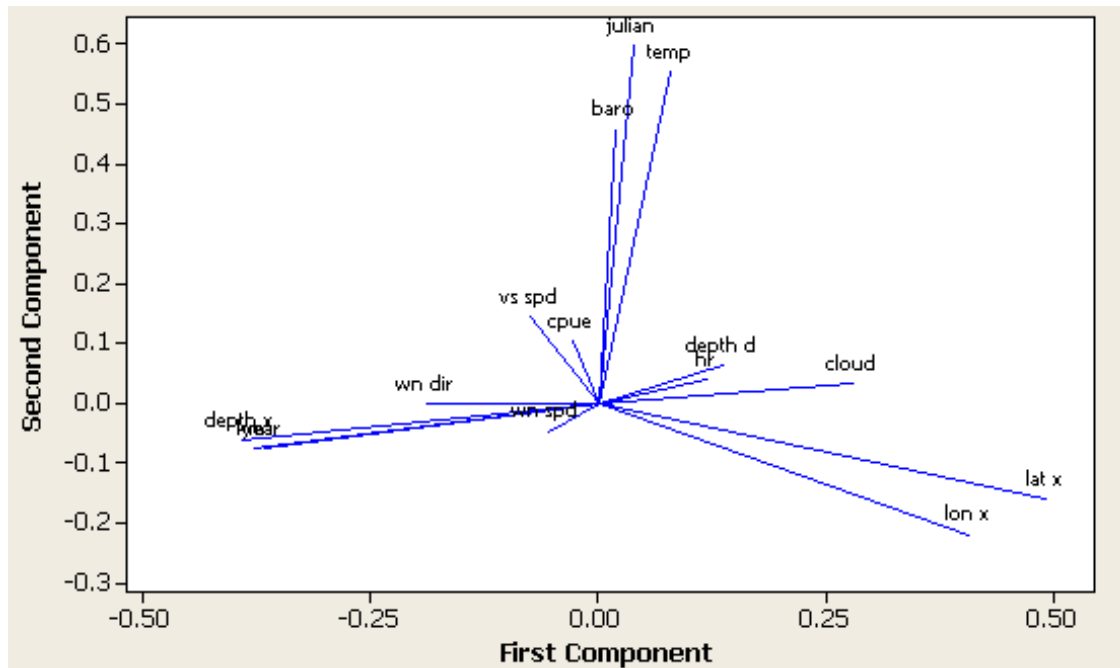
**Figure 2.10.** Mean ( $\pm$  95% C.I.) vessel towing speed during the regional trawl survey.



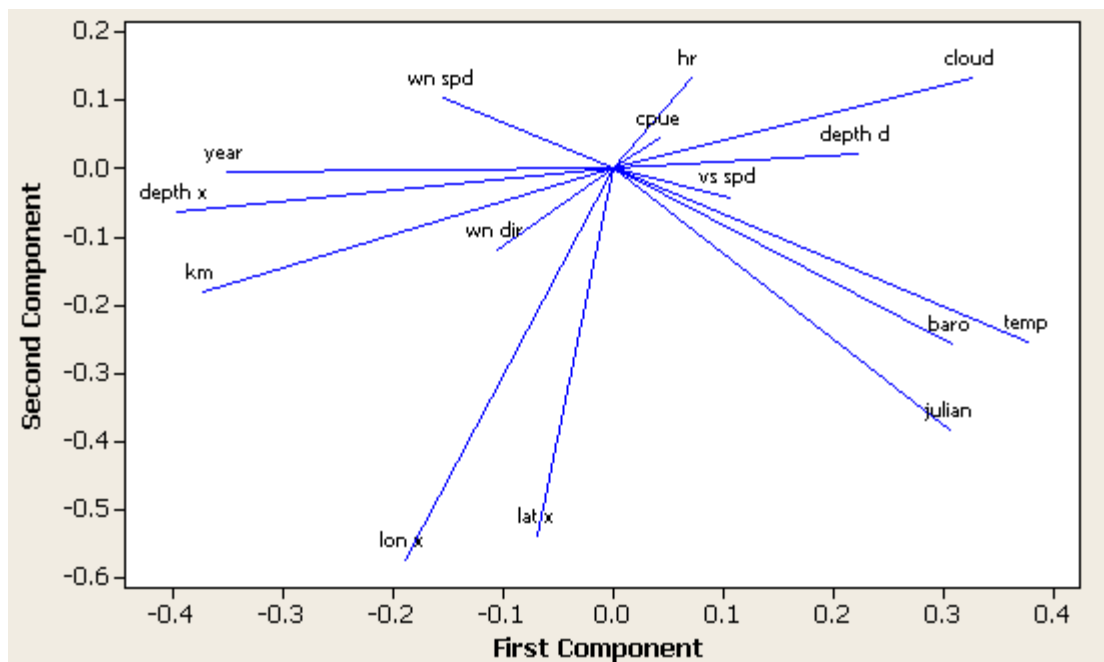
**Figure 2.11.** PCA correlations between CPUE and 14 temporal, spatial, hydrographic and meteorological factors between St. Augustine, FL, and Brunswick, GA (2000-2003, 2008).



**Figure 2.12.** PCA correlations between CPUE and 14 temporal, spatial, hydrographic and meteorological factors between Brunswick, GA, and Savannah, GA (2000-2003, 2008).



**Figure 2.13.** PCA correlations between CPUE and 14 temporal, spatial, hydrographic and meteorological factors between Savannah, GA, and Charleston, SC (2000-2003, 2008).



**Figure 2.14.** PCA correlations between CPUE and 14 temporal, spatial, hydrographic and meteorological factors between Charleston, SC, and Winyah Bay, SC (2000-2003, 2008).

### **Chapter 3 Loggerhead catch rates in two historically studied shipping entrance channels (Charleston, SC; Port Canaveral, FL) in the Southeastern USA.**

#### **Introduction**

The need to observe sea turtles in the water is “self-evident” (Epperly, 2000). However, simple observation of sea turtles is not entirely sufficient, given the need to collect biological sampling to assess turtle health and demographic composition (Braun-McNeill et al., 2007). Furthermore, not all in-water collection techniques are equal, and each offers an inherent suite of attributes and disadvantages with respect to logistical constraints (cost, labor, spatial coverage) and the ability to interpret trends in the data with a high degree of statistical confidence. Generally speaking, variability in catch rate data increases as spatial and temporal coverage increases; however, broader coverage areas also equate to greater opportunities for randomized sampling and ultimately, achieving the goal of representative data. In the absence of sufficient recaptures of tagged individuals, in-water surveys enable assessment of catch rate trends but not abundance (Bjorndal and Bolten, 2000). Among in-water collection techniques in murky coastal waters more than 5m deep, trawl surveys remain the gold standard.

Trawl surveys to collect sea turtles in shipping entrance channels characterize the majority of efforts expended in the Southeastern USA (SE USA) since the 1970's. Trawling in shipping channels has been conducted for various reasons, principally to evaluate seasonal abundance of sea turtles with emphasis on determining windows of opportunity for conducting channel dredging (Richardson and Hillstead, 1979; Van Dolah and Maier, 1993; Dickerson et al., 1995). In conjunction with dredging operations, trawling also enables physical removal of sea turtles from channels in advance of project startup (Joyce, 1982). The evaluation of gear efficiency and sea turtle catchability, two crucial parameters for establishing relative abundance estimates, have also made use of shipping channels to increase the probability of sea turtle collection (Butler et al., 1987; Standora et al., 1993a,b). Lastly, trawling in shipping channels has also been used extensively to collect turtles for behavioral studies (Keinath et al., 1992, 1995; Nelson, 1996; Nelson et al., 1987) and health assessments (Lutz and Dunbar-Cooper, 1987; Bolten et al., 1994).

Given historic sampling efforts, continued sea turtle studies in shipping channels provide some of the longest observational records for assessing changes in catch-per-unit effort. The Port Canaveral, FL, channel was extensively studied between the late 1970's through the mid 1980's (Carr et al., 1980; Henwood, 1987b) and again in the early 1990's (Dickerson et al., 1995). Data for the Charleston, SC, channel primarily dates to the early 1990's (Van Dolah and Maier, 1993; Dickerson et al., 1995; Keinath et al., 1995). The first objective of this chapter is to report catch rates observed in these channels during sampling in spring and summer of 2004-2007. The second objective is to evaluate a suite of factors which may have influenced observed catch rates. In Port Canaveral, adult male loggerhead (*Caretta caretta*) sea turtles were also collected for physiological research (Blanvillain et al., 2008) and at both locations temporal and spatial distribution patterns were also studied, with data presented in Chapters Four (satellite telemetry with juvenile loggerheads from the Charleston, SC, channel) and Five (satellite telemetry with adult male loggerheads from the Port Canaveral, FL, channel), respectively.

## Methods

### *Data collection*

Turtle and data collection are described in the General Methods section of this report. Twelve sampling events (i.e., paired net station) in the Charleston, SC, shipping channel and three sampling events in the Port Canaveral, FL, shipping channel were removed from consideration in analyses due to gear fouling. Assignment to 10-cm size bins for three injured turtles (Charleston) was achieved using straight-line carapace width. Minimum straight-line carapace length for two adult males >95.0cm (caliper maximum in 2006) was approximated using minimum curved carapace length.

Station metadata (vessel speed; surface water temperature; wave height; barometric pressure; and wind speed and direction) were recorded for each sampling event as described in Chapter 2. For Charleston, SC, water level data (15 min intervals) were obtained from the United States Geological Survey (USGS) for Fort Sumter, SC (Station 02172100). Water level data were managed in MS Access and queries were used to determine the following metrics associated with each sampling event: tide stage (ebb, flood); water level difference (ft) between high and low tide; and the percent of tide stage expired at the time of sampling.

By-catch for each sampling event were identified to lowest possible taxon along with quantity per net by individual counts or estimated counts, with estimation being the prevalent method when turtle catch rates were high. The following by-catch species or species groups were analyzed: horseshoe crabs; blue crabs; stone crabs; miscellaneous crabs; whelks; cannonball jellyfish; and miscellaneous jellyfish (including Ctenophora). Similar to loggerhead CPUE (Chapter 2 and this chapter), by-catch CPUE was calculated as specimens per 30.5m net-hr.

### *Data analyses*

Standardized CPUE was computed for loggerhead sea turtles at both sampling locations per the methods described in Chapter 2 of this report. Spatial and temporal analyses of CPUE data were performed with Kruskal-Wallis rank testing (MiniTab®; Minitab, Inc.). Inter-annul variability among station metadata was also statistically tested with Kruskal-Wallis rank testing (MiniTab®; Minitab, Inc.).

Principal Components Analysis (PCA) was performed in MiniTab® (Minitab, Inc.) to evaluate relationships between CPUE for each sampling event and a suite of factors. PCA was only conducted for data from the Charleston, SC, channel, due to temporal variability in sampling, a subsequent volume of replicate observations, and generally (2004-2006) randomized sampling. In addition to CPUE, 18 factors were evaluated and consisted of three sampling metadata factors (station sampling block, time of day and chronological sampling period); eight hydrographic and meteorological factors (wind speed and direction, wave height, water temperature, barometric pressure, and tide stage (% of tide stage elapsed at the time at the start of each sampling event), direction (1=flood, 2=ebb), and amplitude (difference in ft between high and low tide); and seven by-catch species or species groups.

## Results

### *CPUE, Charleston, Loggerhead*

Two hundred sixteen loggerhead turtles (including seven collected twice during this study) were collected in 409 sampling events conducted in the Charleston, SC, shipping entrance channel (Table 3.1). Two Kemp's ridley (*Lepidochelys kempii*) and one green (*Chelonia mydas*) sea turtle were also collected, all in 2004.

Sampling frequency among station blocks (A, B, D) was not significantly different (2004-2006) in May (Chi-sq <sub>$\alpha=0.05, df=4$</sub> ,  $X=0.48$ ,  $p=0.975$ ) or August (Chi-sq <sub>$\alpha=0.05, df=2$</sub> ,  $X=1.62$ ,  $p=0.446$ ). Forty-two percent of stations in June 2004 occurred in the "A" block compared to only 29% of stations in each of the "B" and "D" blocks. Sampling during 2007 primarily focused on the "D" block (93% of stations in May, 63% of stations in August), with all remaining stations sampled from the "B" block and no stations sampled from the "A" Block.

Loggerhead CPUE was significantly different (K-W,  $df=7$ ,  $p=0.001$ ) among eight sampling periods between May 2004 and August 2007. Greatest mean CPUE (3.9 loggerheads per 30.5m net-hr) occurred in May 2004 and lowest mean CPUE (0.5 loggerheads per 30.5m net-hr) occurred in August 2005 (Figure 3.1). Significant differences among six remaining sampling periods were not detected (K-W,  $df=5$ ,  $p=0.991$ ), during which time mean CPUE ranged from 1.3 (August 2007) to 2.9 (May 2006) loggerheads per 30.5m net-hr.

Spatial differences in CPUE were noted with respect to position within the shipping channel, with greatest catch rates associated with furthest distance seaward from the harbor entrance. During May 2004, CPUE increased significantly (K-W,  $df=2$ ,  $p=0.017$ ) between the A, B and D blocks, respectively. CPUE also increased significantly (K-W,  $df=2$ ,  $p<0.001$ ) between the A, B and D blocks, respectively, during June and August 2004 and May 2005 and 2006. During August 2005, CPUE was significantly greater in block D (K-W,  $df=2$ ,  $p=0.011$ ); however, no significant difference in CPUE was observed between blocks A and B (K-W,  $df=1$ ,  $p=0.905$ ). Spatial effects were not tested for sampling in 2007 due to non-systematic sampling.

### *CPUE, Charleston, by-catch*

Blue crabs and miscellaneous jellyfish were the most frequently observed by-catch groupings, and both were most prevalent in May 2006 (Figure 3.2); however, observations for May 2004 may be under-estimated due to high turtle collection (and less robust recording of by-catch data). Blue crabs were more prevalent than horseshoe crabs in all sampling periods. Miscellaneous crabs were prevalent in May 2004, but similar to stone crabs and whelks, miscellaneous crabs were rarely observed during the other seven sampling periods. Cannonball jellyfish were most prevalent in May 2006; however, mean catch was only 0.066 specimens per 30.5m net-hr.

### *CPUE, Canaveral, Loggerhead*

One hundred fifty-eight loggerhead collections occurred during 58 sampling events in the Port Canaveral, FL, shipping channel during April 2006 and 2007 (Table 3.2). Twenty-seven percent ( $n=42$ ) of collections were adult male loggerheads, of which one escaped, two were within-year recaptures, and one was a between-years recapture. Six percent ( $n=9$ ) of loggerheads were adult



females. Sixty-seven percent ( $n=107$ ) of loggerheads were juveniles, of which all but 35 were released without processing; thus, some may have been captured more than once. No significant difference was detected in the ratio of adult males to adult females and juveniles between the three sampling periods (Chi-sq $_{\alpha=0.05, df=4}$ ,  $X=1.62$ ,  $p=0.277$ ).

No significant differences in CPUE were detected among sampling periods for all life history stages combined (K-W,  $df=2$ ,  $p=0.446$ ); for adult males (K-W,  $df=2$ ,  $p=0.601$ ); for adult females (K-W,  $df=2$ ,  $p=0.942$ ); or for juveniles (K-W,  $df=2$ ,  $p=0.097$ ).

Sampling was conducted in four blocks between buoys “9” (inshore) and “1” (offshore); however, sampling effort was not evenly distributed. Fifty percent of sampling occurred between buoys “7” and “5”; 36% of sampling between buoys “5” and “3”; 12% of sampling between buoys “3” and “1”; and only one sampling event (2%) between buoys “7” and “9”.

Significant differences in CPUE (for all life history stages combined) were detected among three buoy blocks (7 to 5; 5 to 3; 3 to 1; K-W,  $df=2$ ,  $p=0.035$ ). Greatest median CPUE occurred closer to shore and decreased progressively seaward. Median CPUE was greater between buoys “7” and “5” (9.2 loggerheads per 30.5m net-hr) than between buoys “5” and “3” (6.2 loggerheads per 30.5m net-hr); however, differences were not significant (K-W,  $df=1$ ,  $p=0.053$ ). Significant differences in CPUE were not detected for adult males among blocks (K-W,  $df=2$ ,  $p=0.435$ ). Similarly, adult female CPUE was not statistically different (K-W,  $df=2$ ,  $p=0.120$ ) among blocks; however, two-thirds of females ( $n=6$  of 9) were collected statistically more frequently between buoys “7” and “5” than between buoys “5” and “3” (K-W,  $df=1$ ,  $p=0.041$ ). Juvenile CPUE was significantly greater between buoys “7” and “5” (K-W,  $df=2$ ,  $p=0.012$ ) than the other two buoy blocks, which were not significantly different from each other (K-W,  $df=1$ ,  $p=0.885$ ).

#### *Hydrographic and meteorological conditions, Charleston*

All hydrographic and meteorological parameters measured at sea were significantly different among months and years (Table 3.3). Comparing May sampling periods among years, May 2004 was associated with the warmest water temperatures and highest barometric pressure, but lowest wind speeds. Wind direction in May 2004 (and 2005) was from the SE, compared to SSW during May 2006 and 2007. Comparing August sampling periods, August 2005 was associated with warmest water temperature, lowest wind speed, and wind direction from the SSE as opposed to from the E as in August 2004 and 2007.

#### *Hydrographic and meteorological conditions, Canaveral*

Significant differences among sampling periods (Table 3.4) were not detected for surface water temperature (K-W,  $df=2$ ,  $p=0.413$ ), wind speed (K-W,  $df=2$ ,  $p=0.412$ ) or wind direction (K-W,  $df=2$ ,  $p=0.238$ ). However, significant differences among sampling periods were detected in barometric pressure (K-W,  $df=2$ ,  $p<0.001$ ) and wave height (K-W,  $df=2$ ,  $p<0.001$ ), with greatest wave heights (2-6 April) and barometric pressure (25-29 April) observed in 2007. Significant differences were also noted in vessel speed among sampling periods, with slightly greater vessel speeds (2.9 vs. 2.8 kts) observed in conjunction with greatest wave heights.

### *Principal components analysis, Charleston*

Data for all 19 factors were available for 89% ( $n=366$  of 409) of sampling events. Individual components each accounted for between 1.4% (PC19) and 11.6% (PC1) of variance (Table 3.5). Strongest relationships (all positive) were observed for the first four components (Figure 3.3); however, these components only accounted for 37.5% of total variance. Tide stage was most closely aligned with CPUE, with association among higher coded (i.e., “ebb”) tide stages. Percent of tide elapsed was the next most closely aligned with CPUE, followed by sampling block (coded landward to seaward) within the channel. Wind speed had the fourth strongest association with CPUE. Inclusion of the fifth (temperature) and sixth (wind speed) components increased total variance accounted for 51.8%. Strong inverse relationships with CPUE were not noted; however, strong inverse relationships were noted between barometric pressure and sampling period, water temperature and blue crab collection, and vessel speed and collection of miscellaneous crabs and jellyfish.

### **Discussion**

Catch rates in the Charleston, SC, shipping channel between 2004 and 2007 exceeded catch rates observed during the same temporal window between 1991 and 1992, despite variability observed during the eight sampling periods between 2004 and 2007. Van Dolah and Maier (1993) reported a peak catch rate of approximately 2.7 turtles per trawl in July 1991, which Maier et al. (2004) converted to 0.35 loggerheads per 30.5m net-hour. However, half ( $n=6$  of 12) of those collections occurred at night (Van Dolah et al., 1992), when significantly greater catch rates were reported (Van Dolah and Maier, 1993). Thus, the daytime catch rate for July 1991 was actually 0.18 loggerheads per 30.5m net-hr, which is 64% less than our lowest (August 2005) monthly mean CPUE of 0.50 loggerheads per 30.5m net-hr. Dickerson et al. (1995) collected six loggerheads in 574 minutes of trawling during the peak of summer 1992 sampling, which equates to 0.48 loggerheads per 30.5m net-hr and is similar to our August 2005 catch rate.

Lower recapture rates observed in 2004-2007 may also suggest an increase in the loggerhead population utilizing the shipping channel. Van Dolah and Maier (1993) recaptured seven of 53 (13%) loggerheads tagged and released during their 16 month survey. Dickerson et al. (1995) collected 45 loggerheads, of which four (9%) were recaptured during their 11 month survey and four previously tagged loggerheads (three by Van Dolah and Maier, 1993) were also recaptured. In contrast, only seven (3.2%) of 216 loggerheads tagged during eight sampling periods between 2004 and 2007 were recaptured, four of which were recaptured within a two-week sampling period. Similarly, only two loggerheads tagged by other studies (both by our trawling efforts off SC in 2000-2003) were collected. Low recapture rates in 2004-2007 were double the recapture rate reported for randomized trawling off SC, GA and northern FL (Maier et al., 2004), perhaps owing to the restricted sampling area and seasonal site fidelity for this location (Chapter 4).

Loggerheads in both shipping channels exhibited clustered rather than randomized distributions, which were generally consistent with historic observations at both locations. Loggerhead collections in the Charleston, SC, channel were predominantly in the “D” sampling block (furthest offshore), which was substantiated in Principal Components Analysis. Aggregation of loggerheads in the “D” block was first reported by Van Dolah and Maier (1993), during monthly trawl surveys between September 1990 and November 1991. Conversely, sampling in the

Charleston channel between September 1991 and March 1993 indicated no statistical difference in spatial distribution (Dickerson et al., 1995). Lack of spatial differences during the 1991-1993 sampling period may stem from sampling the center of the channel to avoid “edge effects” (Dickerson et al., 1993), whereas edges were sampled by Van Dolah and Maier (1993) and the current study. Furthermore, Dickerson et al. (1995) sampled fewer ( $n=3$ ) and longer (3km vs. 1.5km) stations than those sampled by Van Dolah and Maier (1993) and the current study; thus, fine-scale habitat differences should have been less discernible. Greatest loggerhead CPUE in the Port Canaveral, FL, channel occurred between buoys “7” and “5”; however, relatively high CPUE also occurred in the adjacent (and seaward) trawling block. Similar to Dickerson et al. (1995), our trawling efforts in the Port Canaveral, FL, channel occurred along the central axis of the channel. In both studies, greatest loggerhead catch rates occurred between the landward-most and seaward-most sampling blocks, and subtle differences between studies likely stem from delineation of sampling block boundaries. Standora et al. (1993a) also concentrated sampling in the vicinity of buoy “5”, based on an earlier report (Bolten and Bjorndal, 1990) of relatively high loggerhead abundance.

Limited sampling efforts in the Port Canaveral channel suggest comparable catch rates of adult male loggerheads between 1992 and 2006-2007. During this study, 42 adult male loggerheads were collected during 18.8 standardized trawl hours, or 2.2 adult male loggerheads per 30.5m net-hr. In comparison, Dickerson et al. (1995) collected 18 adult male loggerheads during 28 sampling events (mean tow = 25.92 min) in April 1992 (peak annual collection of adult males), or 1.1 adult male loggerheads per 30.5m net-hour, provided that release locations in Table C1 of Dickerson et al. (1995) denote the number of adult male loggerheads collected in a given trawling event. Although catch rates in 2006-2007 were not statistically different from 1992, if the assumption regarding Table C1 is correct, adult male loggerheads in April 1992 were only collected in 39% ( $n=11$  of 28) of sampling events, with more than two adult male loggerheads at a time only collected for a single sampling event. Similar frequency of collection of adult male loggerheads occurred in April 2006-2007 (45%, 26 of 58 events); however, collection of more than two adult male loggerheads at a time was much more common (19%, 5 of 26 events).

Catch rates for adult females and juveniles in the Port Canaveral channel were greater in April 2006-2007 than in April 1992. In April 2006-2007, nine adult females and 107 juvenile loggerheads were collected, compared to collection of just one adult female and 11 juveniles (one of which was previously tagged) in April 1992 (Dickerson et al., 1995). Henwood (1987b) and Dickerson et al. (1995) both report strong seasonal shifts between loggerhead life history stages and sexes encountered in the Port Canaveral channel; juveniles represent more than three-quarters of all loggerheads collected between August and March, with adult males prevalent between March and May and adult females prevalent between May and July. Henwood (1987b) also noted that the apparent arrival of one life history stage coincided with the apparent emigration of other life history stages, primarily expressed as a mass juvenile egress from the channel to accommodate adults. Thus, the abundance of juvenile loggerheads in April 2007 (particularly in late April) may have indicated opportunistic use of the channel by juveniles as adult males transitioned away from the channel, but before adult females arrived en masse. Conversely, hydrographic conditions (as suggested by higher barometric pressure and winds from the SE) in April 2007 (particularly late April) may have resulted in more favorable

conditions than those encountered in April 2006 for encouraging greater use of the channel by all loggerhead life stages, including adult females which may seek inter-nesting refuge in the channel (Meylan et al., 1983). Components analysis for hydrographic and meteorological relationships with loggerhead CPUE was not conducted for Port Canaveral, FL, due to the relatively low frequency and limited temporal scope of sampling events.

Components analysis for loggerhead collections in the Charleston, SC, shipping channel revealed insightful observations with regard to tidal cycles and occurrence of loggerheads in the channel, which has potentially profound implications for management and policy. Although none of the 18 factors in addition to loggerhead CPUE accounted for a large percentage of the variance individually, strongest associations were noted between loggerhead CPUE and tide stage, percent of the tide stage elapsed, and location within the channel. Specifically, loggerhead CPUE was generally greatest furthest offshore towards the end of ebb tide stages. Dickerson et al. (1995) also noted catch rates that were more than double (0.326 vs. 0.141; Table 2) during ebb tide stages than flood tide stages in the Charleston channel; however, statistical differences were not reported. In Chesapeake Bay, loggerheads are documented to ‘ride the tide’ in natural channels of tributaries (Byles, 1988). Furthermore, most loggerheads ( $n=19$  tracks) manually monitored in the vicinity of the Charleston channel during spring 1993 spent less than 15% of the time within the confines of the channel; instead, more time was spent on the adjacent shoals, particularly near the entrance to the channel (Keinath et al., 1995). Acoustic tracking studies were attempted during this research in 2004, but were terminated in lieu of satellite telemetry techniques; however, limited observations (Maier et al., 2005) in spring and summer 2004 were consistent with observations previously reported by Keinath et al. (1995).

In addition to the potential that the “D” block represents a prime foraging location, aggregation of loggerheads in the “D” block may also reflect a ‘deposition effect’, whereby loggerheads are swept seaward in the channel to this location. If true, loggerheads may be most susceptible to boat-strike interactions with shipping traffic in this vicinity of the channel, particularly at the end of ebb tide stages. Indeed, a loggerhead collected freshly dead following obvious interaction with a large boat propeller in May 2006 (Arendt et al., 2007) was collected from the “D” block; however, the tide stage was mid-flood. Wind speed at the time of collection of this turtle was 12kts, and higher wind speeds exhibited the fourth strongest relationship with loggerhead occurrence in the Charleston, SC, shipping channel. Therefore, additional research to better understand the nature of occupancy of the channel by loggerheads is highly advisable. Although we abandoned acoustic telemetry studies due to logistical difficulties, these recent findings collectively suggest that continuous monitoring for acoustically tagged loggerheads at selected locations throughout the channel would be incredibly beneficial for documenting utilization patterns, even if continuous monitoring of the entire channel could not be achieved. Time series data from continuous monitoring studies have the potential to document occupancy patterns for specific areas of the channel, as well as direction of movement within the channel; thus, the potential to develop predictive indices for fine-scale determination of periods when loggerheads were most likely to occupy the channel also exists. As such, elucidation of the factors which may influence loggerhead occupancy of shipping channels could provide a strong basis for developing management policy decisions to mitigate vessel strike interactions, a line item recovery objective of the NW Atlantic Loggerhead Recovery Plan (NMFS & USFWS, 2008).

**Table 3.1.** Summary of sampling events and sea turtle catch in the shipping entrance channel to Charleston, SC (2004-2007).

Year	Month	Start	End		N events	A	B	D		<i>C. caretta</i>	<i>L. kempi</i>	<i>C. mydas</i>
2004	May	05/11	05/19		33	13	10	10		46 (1 <sup>^</sup> )		
2005	May	05/09	05/20		70	30	20	20		36 (2 <sup>#</sup> )		
2006	May	05/15	05/26		69	29	20	20		45 (1 <sup>^</sup> )		
2007*	May	05/21	05/22		15	0	1	14		6 (1 <sup>#</sup> )		
2004	June	06/14	06/25		69	29	20	20		56 (1 <sup>^</sup> )	1	1
2004	August	08/23	09/01		43	14	15	14		16 (1 <sup>^</sup> )	1	
2005	August	08/08	08/19		88	38	23	27		11		
2007*	August	07/31	08/01		22	0	8	14		7		
*targeted sampling					Total	409	153	117	139	223	2	1

<sup>^</sup>within-year recapture

<sup>#</sup> between-year recapture

**Table 3.2.** Summary of sampling events and sea turtle catch in the shipping entrance channel to Port Canaveral, FL (2006-2007).

2006		Trawling Block (Channel Buoys #'s)					Total Loggerheads Caught			
Date	Events	1 to 3	3 to 5	5 to 7	7 to 9		N Cc	Adult M	Adult F	Juv
17-Apr	2	0	1	1	0		8	4	0	2 (1*, 1 <sup>^</sup> )
18-Apr	5	2	3	0	0		5	2	2	1
19-Apr	1	0	1	0	0		5	3	0	1 (1*)
20-Apr	2	0	0	1	1		5	2	0	3
21-Apr	3	1	1	1	0		4	1	0	3

2007		Trawling Block (Channel Buoys #'s)					Total Loggerheads Caught			
Date	Events	1 to 3	3 to 5	5 to 7	7 to 9		N Cc	Adult M	Adult F	Juv
2-Apr	3	1	2	0	0		6	4	1	1
3-Apr	4	0	4	0	0		14	4	0	3 (7*)
4-Apr	5	0	4	1	0		8	1	1	5 (1*)
5-Apr	4	0	1	3	0		18	3	0	4 (11*, 1 <sup>^</sup> )
6-Apr	6	0	3	3	0		13	1	1	3 (9*)

2007		Trawling Block (Channel Buoys #'s)					Total Loggerheads Caught			
Date	Events	1 to 3	3 to 5	5 to 7	7 to 9		N Cc	Adult M	Adult F	Juv
25-Apr	5	1	2	2	0		29	4 (1*)	2	6 (21*, 1 <sup>^</sup> )
26-Apr	4	0	1	3	0		12	3	0	2 (7*)
27-Apr	8	2	3	3	0		12	3	1	1 (7*)
28-Apr	2	0	1	1	0		12	5 (2 <sup>^</sup> , 1*)	0	4*
29-Apr	4	0	2	2	0		8	3	1	4*

\* = escaped or not processed

# = dead

<sup>^</sup> = recaptured

**Table 3.3.** Summary of statistical testing for station metadata (vessel speed, hydrographic and meteorological data) during sampling in the Charleston, SC, shipping channel (2004-2007).

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Vessel Speed	N obs	35	68	68	15	68				42	88		22
	Median	2.7	2.8	2.8	2.9	2.7				3.0	2.8		2.9
	SD	0.1	0.2	0.3	0.1	0.1				0.2	0.2		0.2

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.000\*

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Water temperature	N obs	35	70	69	15	69				43	88		22
	Median	24.7	22.6	22.5	22.2	27.0				26.6	28.5		27.0
	SD	0.7	1.1	1.3	0.3	0.6				0.6	0.6		0.3

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.000\*

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Wave height	N obs	35	70	68	15	69				43	88		22
	Median	0.3	0.3	0.3	0.2	0.3				0.3	0.3		0.3
	SD	0.2	0.3	0.2	0.1	0.2				0.1	0.2		0.2

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.008\*

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Barometric pressure	N obs	35	70	69	15	69				43	88		22
	Median	1025.2	1014.3	1009.0	1019.8	1015.6				1017.5	1017.4		1012.3
	SD	1.7	3.0	3.8	1.4	2.6				1.9	1.3		1.2

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.000\*

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Wind speed	N obs	34	61	65	15	69				41	66	22	
	Median	8.0	10.0	10.0	10.0	15.0				10.0	7.0	12.0	
	SD	2.7	4.3	3.7	3.8	3.6				3.7	3.1	2.9	

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.000\*

Parameter	Metric	May				June				August			
		2004	2005	2006	2007	2004	2005	2006	2007	2004	2005	2006	2007
Wind direction	N obs	34	61	65	15	69				41	66		22
	Median	135	135	203	203	203				90	158		90
	SD	25	71	91	90	61				85	76		46

K-W (month), df=2, p=0.000\*

K-W (year), df=3, p=0.005\*

**Table 3.4.** Summary of statistical testing for station metadata (vessel speed, hydrographic and meteorological data) during sampling in the Port Canaveral, FL, shipping channel (2006-2007).

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Vessel Speed	N obs	13		22		22
	Median	2.8		2.9		2.8
	SD	0.1		0.1		0.1

K-W (month), df=2, p=0.000\*

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Water temperature	N obs	13		22		23
	Median	23.8		23.5		23.6
	SD	0.4		0.5		0.3

K-W (month), df=2, p=0.413

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Wave height	N obs	13		22		23
	Median	0.2		0.6		0.3
	SD	0.1		0.1		0.1

K-W (month), df=2, p=0.000\*

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Barometric pressure	N obs	13		22		22
	Median	1016.7		1015.1		1021.8
	SD	2.5		4.3		1.0

K-W (month), df=2, p=0.000\*

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Wind speed	N obs	13		22		23
	Median	4.0		10.0		10.0
	SD	5.0		2.4		2.7

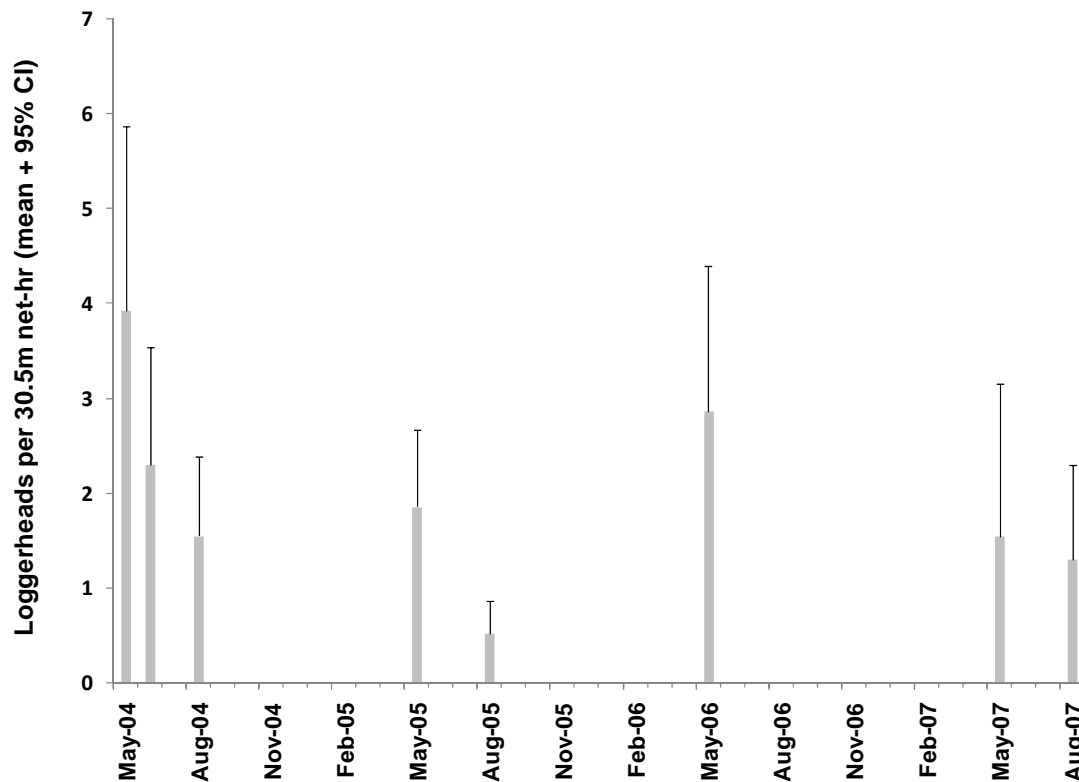
K-W (month), df=2, p=0.412

Parameter	Metric	4/17/06 to 4/21/06		4/2/07 to 4/6/07		4/25/07 to 4/29/07
Wind direction	N obs	13		22		23
	Median	270		158		158
	SD	116		111		76

K-W (month), df=2, p=0.238

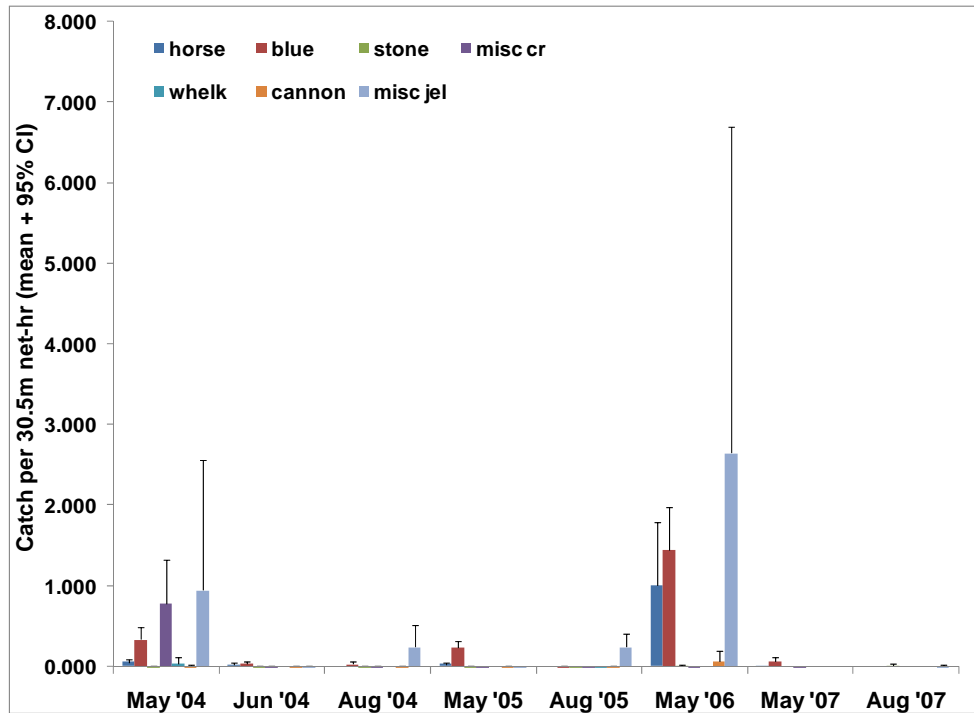
**Table 3.5.** Eigenvalues and variance distribution determined by Principal Components Analysis for loggerhead CPUE plus 18 additional factors in the Charleston, SC, shipping channel.

Metric	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Eigenvalue	2.2027	1.8874	1.5917	1.446	1.4364	1.253	1.1173
Proportion	0.116	0.099	0.084	0.076	0.076	0.066	0.059
Cumulative	0.116	0.215	0.299	0.375	0.451	0.517	0.576
Metric	PC8	PC9	PC10	PC11	PC12	PC13	PC14
Eigenvalue	1.092	0.9917	0.9191	0.9058	0.8438	0.7372	0.6261
Proportion	0.057	0.052	0.048	0.048	0.044	0.039	0.033
Cumulative	0.633	0.685	0.734	0.781	0.826	0.864	0.897
Metric	PC15	PC16	PC17	PC18	PC19		
Eigenvalue	0.5071	0.4648	0.3678	0.338	0.272		
Proportion	0.027	0.024	0.019	0.018	0.014		
Cumulative	0.924	0.949	0.968	0.986	1		

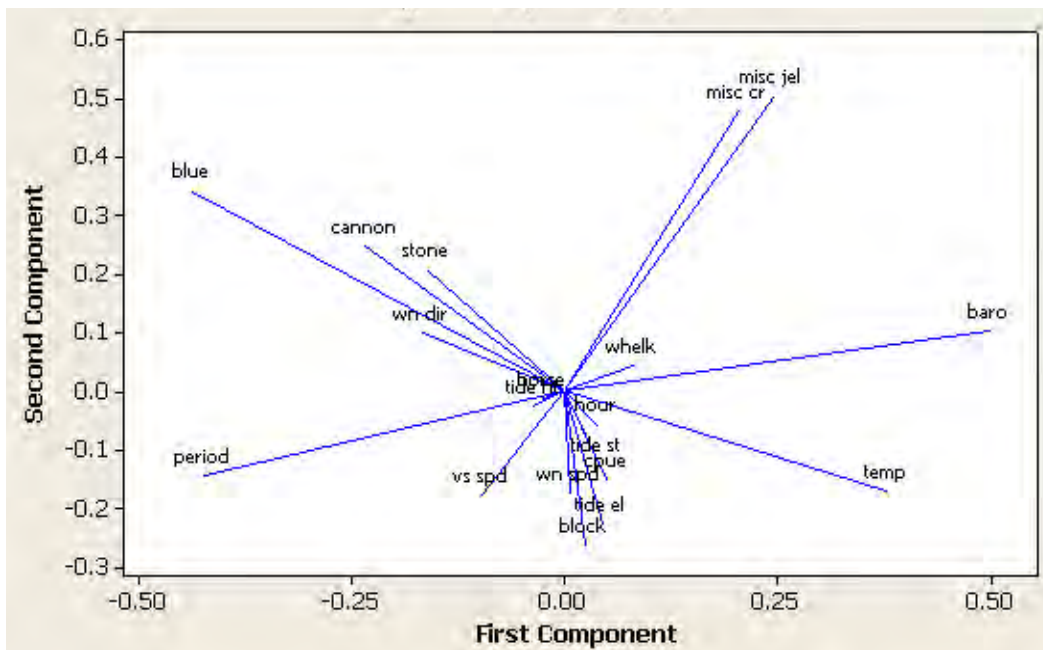


**Figure 3.1.** Loggerhead CPUE (turtles per 30.5m net-hr) for randomized (2004-2006) and targeted (2007) sampling in the Charleston, SC, shipping entrance channel.





**Figure 3.2.** Standardized catch (mean  $\pm$  95% CI per 30.5m net-hour) for selected by-catch groupings collected during sampling in the Charleston, SC, shipping channel (2004-2007).



**Figure 3.3.** Relative importance of 18 factors (temporal and spatial characteristics of sampling plus meteorological, hydrographic, and by-catch factors) to the observed loggerhead CPUE in the Charleston, SC, shipping channel (2004-2007).

## Chapter 4 Seasonal distribution and activity of juvenile loggerheads near Charleston, SC.

### Introduction

Management and protection of highly *k*-selected marine species such as sea turtles is ineffective without the availability of long-term monitoring and reliable data collection. For example, in-water populations for loggerhead (*Caretta caretta*) sea turtles in the Northwest (NW) Atlantic must increase for at least 50 years before recovery can be declared (NMFS and USFWS, 2008). Although data sets older than 50 years are available that document frequency of loggerhead nesting (Caldwell et al., 1959) and in-water occurrence (Bullis and Drummond, 1978), critical corollary data sets to establish the probability of sea turtle encounters (Anderson, 2001) may be incomplete, thereby precluding the use of earliest data sets in population assessments. Factors which may influence the mobility of loggerhead sea turtles are especially important, and include response to changes in water temperature (Coles and Musick, 2000), circulation patterns and frontal boundaries (Polovina et al., 2001) and pursuit of mobile prey (Plotkin et al., 1993).

Given natal homing of loggerhead sea turtles (Bowen et al., 2004), heritage may ultimately be the most important determinant of the probability of a loggerhead inhabiting a specific locale. Although the majority of offspring from all known loggerhead nesting colonies in the NW Atlantic Basin (Bowen et al., 2004) utilize the northern foraging ground (FL to the Northeast (NE) USA), only one nesting colony is associated with the shoreline adjacent to this foraging ground; thus, many loggerheads encountered in this region will not be life-long inhabitants. Migration between southern overwintering areas (Carr et al., 1980) and northern estuaries occurs from south to north in the spring (Lutcavage and Musick, 1985) and in the reverse direction in the fall (Shoop and Ruckdeschel, 1989; Morreale and Standora, 2005). Subsequently, the relative abundance of loggerheads in coastal waters off GA and SC fluctuates seasonally as transient turtles traverse through their migratory corridor. Lastly, habitat preferences and local movements also affect the probability of encountering loggerheads. Although random sampling minimizes habitat bias, it is more difficult to minimize the effects of individual movements.

Recapture events for tagged sea turtles document movement patterns and/or site affinity. However, poor recovery rates for tagged sea turtles (Epperly et al., 2007) often hinder population assessments using tag-recapture data. Because the tag-recapture method does not provide data on the location of tagged sea turtles between tag-release and tag-recovery events, this method also does not allow assessment of potential causes of low tag-recovery rates. Plausible explanations for low tag-recovery rates include: tag loss; emigration from the tag-release and subsequent survey zone; mortality of tagged turtles within or outside of the survey zone; and/or an abundance of untagged sea turtles. In contrast to tag and release, satellite-telemetry allows frequent location assessment of tagged sea turtles, and has been widely used to study their temporal and spatial distribution patterns (Godley et al., 2008). In response to low (1.2%;  $n=10$  of 827 loggerhead collections) loggerhead tag-recapture rates for turtles tagged in coastal waters off SC, GA and northern FL (Maier et al., 2004), this study initiated satellite-telemetry studies with juvenile loggerheads collected near Charleston, SC. The first objective was to document frequency of occurrence within the regional trawl survey area. The second objective was to characterize loggerhead temporal and spatial distribution patterns and assess factors of influence.

## Methods

### *Turtle collection, selection, and satellite transmitters*

Loggerhead sea turtles were collected from the Charleston, SC, shipping entrance channel in May (2005-2007), June (2004), and in August (2004-2005, 2007). Loggerheads were opportunistically selected for satellite tagging, provided they displayed no outward signs of poor health and turtles measured between 55cm and 75cm SCLmin. Upon completion of standard turtle data collection (see General Methods section of this report), ST-20 satellite transmitters (Telonics, Inc.; Mesa, AZ) were attached to the carapace in the manner described below.

ST-20 (Model A2020) satellite transmitters were attached directly to the second vertebral scute of the carapace using epoxy (Papi et al., 1997; Polovina et al., 2000). Prior to attachment, barnacles and other organisms were carefully removed with a chisel, and the carapace was sanded, washed with betadine and dried with acetone. A roll of 1.0 cm diameter Sonic Weld™ (Ed Greene & Company; Sparta, TN) was placed around the bottom edge of the transmitter to form a well. Using a caulking gun, Fast Foil™ (Power Fasteners Inc.; New Rochelle, NY) epoxy was applied to the entire bottom surface of the transmitter within the well. Transmitters were then pressed onto the carapace and the epoxy was allowed to set. Loggerheads were released 1-2 h after and in close proximity (<3km) to where they were originally collected.

### *Data management*

Satellite telemetry data consisted of (1) geographic position at each surfacing, (2) transmitter temperature (a surrogate for water temperature) at each surfacing, (3) time (s) of last dive before surfacing and (4) three dive cycle metrics corresponding to the number of dives, mean dive duration (s) and the percent of time submerged during four, six-hour data collection bins per day (0000-5559 UTC; 0600-1159 UTC; 1200-1759 UTC; 1800-2359 UTC). Each geographic position was associated with a CLS America location class (LC: 3, 2, 1, 0, A, B, Z) to establish position accuracy. Accuracy estimates were only provided for location classes “3” (<100m), “2” (100-300m), and “1” (300-1000m). Satellite telemetry data were automatically processed and distributed by CLS America system as daily data e-mails; however, data were initially managed using the Satellite Tracking and Analysis Tool (STAT; Coyne and Godley, 2005). Satellite telemetry data were downloaded from STAT to a relational database (MS Access) on a local area network for evaluation and filtering prior to analyses.

Two STAT filters were applied to all location data, regardless of CLS America location class. The topographic filter removed detection events associated with elevations >0.5m above sea level. The inner turning angle filter removed detection events in which the angle between the previous and successive detection events was <10°; such events represented “spikes” away from other locations which were clustered together or in a linear series during movement periods. Post-topographic- and inner angle-filtered data were imported into MS Excel, and a speed filter (5km/hr) was applied to all data (excluding LC 3, 2, and 1) to remove additional suspect data points. Lastly, data which passed all three filters were graphically examined to remove any remaining suspect data points (generally ephemeral pulses ≥10km). To remove bias for days with numerous detection events for a given satellite-tagged turtle, a single latitude and longitude was computed as the average of all retained latitudes and longitudes for each turtle for each day.

Transmitter sensor data also required extensive filtering. Transmitter temperature records  $>40^{\circ}\text{C}$  and  $<5^{\circ}\text{C}$  were removed. The remaining temperature data were graphically examined to remove any additional outlying points that appeared as “spikes” in the time series. Data for all three dive metrics were automatically removed when the number of reported seconds (mean dive duration multiplied by the number of dives) exceeded the number of possible seconds ( $n=21,600$ ) in a six hour period. The filtered dive metric data were configured from a database table format into a matrix table format (with columns denoting data collection bin) then visually examined for consistency among replicate observations for a given metric for each data collection period. When two or more values for a given dive metric were in agreement, all other deviations were excluded. If only two records were available, values were averaged if the difference between values was  $<15\%$ ; otherwise, both values were removed from the data set. Only one observation per dive metric per data collection bin was utilized to minimize bias associated with the receipt of multiple (and identical) observations for the same data collection bin.

### *Data analysis*

Daily location estimates for satellite-tagged turtles were spatially analyzed using ArcGIS ArcInfo Desktop 9.2 (ESRI, Redlands, CA). Data for two long-term rehabilitated loggerheads (ID#58944 and ID#49618) were excluded; however, data for a third rehabilitated loggerhead (ID#73120) were analyzed as the rehabilitation time for a puncture wound was minimal.

Satellite-tagged loggerheads were classified into two groups: residents which generally remained within 40km off the coast of SC, GA and northern FL, and transients that rapidly emigrated out of the regional trawl survey area (2000-2003) and did not return. Daily location estimates for resident loggerheads were pooled to evaluate spatial distribution as follows: (1) within the trawl survey boundary; (2) between the north and south boundaries, but inshore of the sampling area; (3) between the north and south boundaries, but between the eastern trawl boundary and 20 m depth contour; or (4) outside of the trawl boundary (north, south and/or water depth  $>20$  m). Trawl boundary was established as a perimeter around the sampling universe ( $n=1502$  stations) from which annual stations were randomly selected during 2000-2003 and 2008. A 1km buffer was placed around the trawl boundaries to account for daily location estimates on the boundaries.

Spatial AutoCorrelation (Morans Index) was performed as described in Chapter 3. For each resident loggerhead, activity centers and the (shoreline-adjusted as needed) area encompassing 95% (two standard deviations) of daily location estimates was established for each of four temporal bins. The first temporal bin (November through January) was associated with rapidly declining water temperature and annual minimum water temperature and photoperiod. The second bin (February through April) was characterized by increasing photoperiod, but only slightly increasing water temperature. The third bin (May through July) represented rapidly increasing water temperature and annual maximum water temperature and photoperiod. The fourth bin (August through October) corresponded with a decline in photoperiod, but only a slight decrease in water temperature. In addition to Moran's Index, area (hectares, ha) encompassed by 95% of daily location estimates was standardized by the number of days each turtle was observed during a given temporal bin. Testing for significant differences in day-standardized (for turtles with at least 20 daily location estimates per temporal bin) areas was performed using Kruskal-Wallis analysis of variance by ranks (Minitab 15®; Minitab, Inc.).

Transmitter sensor data for resident and transient loggerheads were analyzed independently. Statistical differences in transmitter temperature and three dive metrics among four temporal bins were tested using Kruskal-Wallis analysis of variance by ranks. Time of day effects for dive metrics were also tested (Kruskal-Wallis) separately for each of four temporal bins.

Principal Components Analysis (Minitab 15®; Minitab, Inc.) was used to evaluate correlations between resident loggerhead location status (within trawl survey area vs. inshore or outside), daily mean values for transmitter sensors (temperature; mean dive duration; number of dives; and percent of time submerged) and 11 additional corollary factors. Daily mean values for air and water temperature, wind speed and direction, and barometric pressure and wave height were computed from hourly data recorded by the Edisto Buoy (#41004) located 41nm SE of Charleston ([www.ndbc.noaa.gov/Southeast/shtml](http://www.ndbc.noaa.gov/Southeast/shtml)). Photoperiod cycle (increasing, decreasing) and day length (h) were determined from sunrise and sunset data for Charleston, SC, obtained from the U.S. Naval Observatory (<http://aa.usno.navy.mil/data>). Julian date (1 to 366), and latitude and longitude for each resident turtle day of observation were also evaluated as factors.

## Results

### *General*

Thirty-four juvenile loggerheads (mean = 64.3cm SCLmin; range = 56.6 to 76.8cm) were satellite-tagged after collection from the Charleston, SC, shipping entrance channel (Table 4.1). Seventy-six percent ( $n=26$ ) of loggerheads were female, 21% ( $n=7$ ) were male and sex could not be determined for one loggerhead. The ratio of females to males was not significantly different among years (Chi-square<sub>df=3,  $\alpha=0.05$</sub> ,  $X=4.025$ ,  $p=0.259$ ). Genetic distribution was dominated by CC-A01 ( $n=20$ , 59%) and CC-A02 ( $n=11$ , 32%), and the ratio between these haplotypes was not significantly different among years (Chi-square<sub>df=3,  $\alpha=0.05$</sub> ,  $X=4.025$ ,  $p=0.259$ ).

Eighty-two percent ( $n=28$  of 34) of satellite-tagged juvenile loggerheads were classified as residents (Table 1). Of the six loggerheads classified as transients, four (ID#57684, ID#64552, ID#73121 and ID#73123) primarily relocated to the middle and outer continental shelf offshore from Long Bay (Winyah Bay, SC, to Cape Fear, NC; Figure 4.1). A fifth loggerhead (ID#64550) was most frequently detected just south of St. Augustine, FL, the southern boundary for the regional trawl survey area. A sixth loggerhead (ID#73114) remained north of Cape Hatteras, NC (except during winter), the northern boundary for the South Atlantic Bight (SAB).

All but two resident loggerheads (ID#49123 and ID#53687), and possibly a third resident (ID#58941) remained on the continental shelf for the entirety of their respective data collection. Movement off the continental shelf for all loggerheads occurred between December and March.

### *Spatial and temporal distributions*

A total of 5,358 daily location estimates were analyzed for spatial statistics. Seventy-eight percent ( $n=4,208$ ) of daily location estimates were affiliated with resident loggerheads. Thirty-three percent (Table 4.2) of daily location estimates were collected between May and July; similar data collection levels occurred in November to January (23%) and May to July (26%); and the least amount of data were collected between February and April (18%).

No significant differences (Moran's Index,  $Z=0.29$ , random) in activity centers were detected among temporal bins (Figure 4.2). Area encompassed (ha) was significantly different among seasons (K-W,  $df=3$ ,  $p<0.001$ ); however, considerable individual variability was also noted (Figure 4.3). Area encompassed by resident loggerheads between May and October was approximately 75% smaller than area encompassed between November and April. Area encompassed between November and January was not significantly different than area encompassed between February and April (K-W,  $df=1$ ,  $p=0.742$ ). Similarly, significant differences in area encompassed between May and July versus between August and October were not detected (K-W,  $df=1$ ,  $p=0.431$ ).

Location of resident loggerheads on the continental shelf was seasonally driven (Figure 4.4). Resident loggerheads frequently occurred within the regional trawl area between May and July ( $n=682$  of 1,102 locations, 62%) and August and October ( $n=842$  of 1,358 locations, 62%). Resident loggerhead distribution between May and July was also characterized by the greatest frequency ( $n=202$  of 314, 64%) of locations inshore of the regional trawl survey area and the lowest frequency ( $n=218$  of 2,040; 10%) of locations outside of the regional trawl survey area.

Distribution of resident turtles inshore or outside of the regional trawl area between May and July reflected individual variability. Three resident loggerheads (ID#49120, ID#57685 and ID#73116) accounted for 58% of all inshore locations. Similarly, 59% of all outside locations were attributed to four loggerheads (ID#57683, ID#57687, ID#57688 and ID#49123). Principal components analysis attributed between 0.1% and 25.8% of total variance to each of 16 factors, with the first three factors accounting for 58.3% of the total variance. Strong correlations were noted between disposition status and dive duration and longitude (Figure 4.5). Strong inverse correlations between these three factors occurred with number of dives, percent of time submerged, Julian date, photoperiod (and stage), and transmitter temperature. Environmental factors represented the last six components and collectively accounted for 6.6% of variance.

Nine resident loggerheads were monitored throughout the winter season and into at least April. Directed return to the vicinity of the Charleston, SC, shipping entrance channel was noted for six loggerheads (Figure 4.6), including one (ID#49122) that was recaptured in the channel in May. Two others (ID#58941, ID#73122) appeared to be en route to return, but tracking ceased before a return could be confirmed. The ninth loggerhead (ID#49123) remained in oceanic waters approximately 1200km east of Delaware Bay where it had over-wintered.

#### *Diving behavior and associated factors*

A total of 145,212 transmitter sensor data points were analyzed. Nearly all ( $n=45,448$  of 45,660) transmitter temperature records were retained. Seventy-five percent of dive metric data were retained for individual data collection bins ( $n=13,753$  to 13,819 of 18,383), representing 72-73% retention of raw data records ( $n=32,937$  to 33,424 of 45,590) for each dive metric parameter.

Monthly transmitter temperature closely mirrored seasonal water temperatures recorded at the Edisto Buoy between 2004 and 2008 (Figure 4.7). In winter, resident loggerheads generally occupied waters cooler than sea surface temperature associated with the Edisto Buoy. Smaller

monthly variation for both residents and transients was also associated with transmitter temperatures than was observed for sea surface temperature.

Loggerhead submergence and surfacing patterns also followed seasonal cycles (Figure 4.8). Loggerheads monitored in 2004 and 2005 generally remained at the sea surface for 5-10% of the time between May and November, but spent 12-32% of the time at the surface between December and April. Seasonal changes in submergence patterns were less pronounced for both resident and transient loggerheads monitored during 2006-2007.

The number of dives (Figure 4.9) and mean dive duration (Figure 4.10) for six-hour data collection bins were inversely related. All loggerheads generally made more and shorter dives between May and October, followed by fewer and longer dives between November and April. Dive frequency in winter 2004-05 was slightly greater than dive frequency in winter 2005-06. Similarly, mean dive duration in winter 2004-05 was slightly less than mean dive duration in winter 2005-06. Diel diving behavior was similar among seasons (Figure 4.11), with longer dives between 0000 and 1159 UTC, followed by shorter dives between 1200 and 2359 UTC. Diel periodicity in dive duration was statistically different (K-W,  $df=3$ ,  $p<0.001$ ) for all seasons and dispositions; however, diel variation was least evident for residents between May and July.

## **Discussion**

Between 2004 and 2008, juvenile loggerheads collected from the Charleston, SC, shipping entrance channel and subsequently monitored by satellite telemetry repeatedly demonstrated high site affinity for continental shelf waters off SC and GA. On average, four of every five juvenile loggerheads remained between the northern and southern boundaries of the regional trawl survey between May and October, with 62% of daily location estimates for these loggerheads occurring within the eastern and western boundaries of the regional trawl survey area as well. Resident loggerheads predominantly over-wintered further offshore from the same latitudes occupied during warm water periods, in such proximity as to preclude detection of spatial differences between seasonal distributions. Among loggerheads that did not initially remain between the northern and southern limits of the regional trawl survey area, five generally remained within 100km of the survey area; four of the five turtles occurred just north of the northern trawl survey boundary and the fifth loggerhead relocated just south of the southern boundary. Only one loggerhead did not remain in the SAB during the warm water months.

Prior to this study, seasonal distribution patterns of juvenile loggerheads in coastal waters south of NC were poorly documented. In contrast to data for more than 30 adult female loggerheads satellite-tagged on nesting beaches in GA (Stoneburner, 1982; GADNR, unpublished data) and SC (SCDNR, unpublished data 2), distributional data for juveniles in the same area consisted of short-term acoustic and/or radio tracking data (Keinath et al., 1992, 1995; Nelson, 1996) and limited satellite telemetry data for three juvenile loggerheads. A rehabilitated juvenile traversed back and forth within 50km of shore between Port Royal Sound, SC, and St. Augustine, FL, between September 1998 and November 1999 (NMFS, unpublished data). Two trawl-caught juveniles were released near Brunswick, GA, in March 1999 (USACOE, unpublished data). One juvenile was tracked for 2.5 months and initially traveled south before traveling north generally within 30km of shore; off the coast of Savannah, GA, this turtle moved offshore as far as 75km, but returned to within 30km of shore and continued north to Charleston, SC, where it was last

observed. The second turtle was tracked for four months and made regular longitudinal movements up to 150km offshore between Cumberland Island, GA, and Savannah, GA. Seasonal distribution patterns for juvenile loggerheads that remained on the continental shelf in this study generally resembled distributional patterns noted for juvenile loggerheads collected from estuarine waters between NC and NY. Loggerheads in this study exhibited both strong site affinity and site fidelity following over-wintering. Van Dolah and Maier (1993) also reported site fidelity for the Charleston, SC, shipping entrance channel, noting that four of their eight recaptures occurred the following spring and summer after over-wintering elsewhere. Rapid return to areas occupied in the previous fall has also been documented for juvenile loggerheads satellite-tagged north (Keinath, 1993; Morreale and Standora 1994; Morreale, 1999; Mansfield, 2006) and just south (McClellan and Read, 2007) of Cape Hatteras, NC. However, an important distinction between this study and previous studies pertains to segregation of shelf over-wintering areas. Most juvenile loggerheads originating from estuaries between NC and NY overwintered off of Raleigh Bay (Cape Lookout to Cape Hatteras, NC) a known winter aggregation area for loggerheads (Epperly et al., 2005b); however, some juveniles in previous studies also over-wintered off central FL (Keinath, 1993; McClellan and Read, 2007). In contrast, no loggerheads in the current study overwintered off central FL and only one loggerhead in the current study over-wintered off Raleigh Bay; perhaps not coincidentally, this same loggerhead was the only one that did not remain within the SAB during warm water periods (and instead alternated between the Delaware and Chesapeake Bays in 2006 and 2007, respectively). Thus, similar to co-occurrence of resident and transient populations for bottlenose dolphins (*Tursiops truncatus*) along the U.S. East Coast (Zolman, 2002), distinct resident loggerhead foraging groups may exist between FL and NY, but which are linked via migration corridors and seasonal movements of transients (Sasso et al., 2006). Mitochondrial DNA analyses also support the suggestion of distinct resident areas (Bowen et al., 2005).

A second over-wintering strategy documented in this study involved departure from the continental shelf to occupy oceanic habitats hundreds of kilometers offshore. Although only two juvenile loggerheads (and possibly a third) in our study actually made this transition, because only 10 total loggerheads were able to be monitored through the winter, the proportion of juvenile loggerheads over-wintering (at least partially) in oceanic habitats is 20-30%. A similar proportion of juvenile loggerheads monitored by satellite telemetry elsewhere on the U.S. Eastern Seaboard have also made this transition (Keinath, 1993; Morreale and Standora 1994; Morreale, 1999; Mansfield, 2006; McClellan and Read, 2007). Transition from shelf to oceanic habitats consistently occurs during the winter with all loggerheads entering the Gulf Stream in the vicinity of Cape Hatteras, NC. Loggerheads may remain in oceanic habitats for a few months (Morreale, 1999) or more than a year (Mansfield, 2006) before returning to the continental shelf. Even if juvenile loggerheads never returned to shelf waters after ‘switching back’ to oceanic habitats, it is unwise to consider oceanic loggerheads “lost” given that the majority of offspring from every known nesting colony in the North Atlantic Basin forage between FL and NY at some point during their lifetime (Bowen et al., 2004). However, as McClellan and Read (2007) point out, loggerheads are considerably more vulnerable to negative interactions with long-line fisheries when utilizing oceanic vs. neritic habitats.



Despite a common collection location and overlap in spatial distributions (which encompassed the shipping channel), loggerheads spent very little time in the actual channel. Seasonal affinity for, but only episodic occurrence within, shipping channels has been documented at multiple locations in the Southeastern US (SE USA) using manual acoustic and/or radio telemetry. Keinath et al. (1995) monitored 19 sub-adult loggerheads near the harbor entrance channel in Charleston, SC, and 12 sub-adult loggerheads near the harbor entrance channel in Savannah, GA, during spring and fall 1993 and noted that 80% ( $n=24$ ) of them spent less than 10% of the time monitored within the confines of the entrance channel. Nelson (1996) monitored 20 juvenile loggerheads near the St. Mary's shipping entrance channel in King's Bay, GA, for 822h in the spring, summer and fall of 1993 and found that loggerheads generally occupied single locations in 10-20m water depths for up to 12h before moving 1-5km along shore to occupy new locations. Loggerheads were generally located  $> 0.5\text{km}$  away from the entrance channel during 90% of survey period, but typically remained within 9km of the channel (Nelson, 1996). Kemmerer et al. (1983) reported similar observations for short-term residence of juvenile loggerheads in the vicinity of the Canaveral shipping channel; data were only collected for 12 loggerheads, but all of them were relocated up to 20 days (the duration of the study) after release. In contrast, Keinath et al. (1992) obtained 138 positional fixes for five juvenile loggerheads during 73.75h of monitoring, during which 95% of positional fixes were distributed within the shipping entrance channel to St. Simon's Sound, GA, in water depths  $> 6\text{m}$ .

Activity centers and area encompassed by loggerheads were highly variable among individuals. Assessments of seasonal area utilized in this study were meant to provide relative comparisons and not precise estimates of home range size; thus, in rare cases of linear distributions within areas, assessments encompassing the span of 95% of daily locations would be over-estimated. Renaud and Carpenter (1994) satellite tagged three juvenile loggerheads in the eastern Gulf of Mexico (GOM) and defined a core area as encompassing 50% of all locations; core areas for two resident loggerheads each monitored for six months were  $89\text{-}98\text{km}^2$ , though overall home range spanned between  $954$  and  $1435\text{km}^2$ . Estimates of home range from manual tracking studies, in which precise locations were able to be obtained, also demonstrate remarkable mobility for even "localized" loggerheads exhibiting high site fidelity. Byles (1988) conducted extensive radio and acoustic telemetry monitoring of juvenile loggerheads ( $n=12$ ) within a  $350\text{km}^2$  portion of the Chesapeake Bay and associated tributaries between 1981 and 1984. Home ranges were estimated to be  $10\text{km}^2$  for 67% of turtles ( $n=8$ ) and up to  $80\text{km}^2$  with a preferred area of  $12\text{km}^2$  within larger home ranges for 33% ( $n=4$ ) of turtles (Byles, 1988). Tidal flow had pronounced influence on local movement patterns, with two-way linear movements in the direction of tide flow generally  $<8\text{km}$  (but up to  $27\text{km}$ ); however, at times loggerheads also remained stationary (Byles, 1988). Avens et al. (2003) also reported strong site fidelity for loggerheads trans-located following capture in pound nets; loggerheads rapidly returned (3-5d) and also remained within areas approximately (based on figure scales)  $10\text{km}^2$ .

Seasonal change in water temperature was the strongest determinant for occurrence of juvenile loggerheads on the inner continental shelf. Between late fall and early spring, juvenile loggerheads remained on the middle to outer continental shelf, in a narrow band of tempered water between the western wall of the Gulf Stream and a 'slug' of cold water that stretched approximately  $40\text{km}$  offshore and which was evident from sea surface temperature (a map layer

feature activated in STAT). Overwintering of loggerheads in this vicinity is consistent with observations from aerial surveys conducted in the early 1980's (Thompson, 1984); thus, our observations do not appear to reflect a recently acquired over-wintering strategy in response to perceivably milder winters. Abrupt increase in water temperature above 17°C in the spring triggered the movement of loggerheads inshore. Similarly, abrupt decrease in water temperature below 17°C in the fall also prompted loggerheads to return offshore. Coles and Musick (2000) utilized satellite sea surface temperature to establish a preferred temperature range of 13-28°C for loggerheads distributed off the coast of NC. Epperly et al. (1995b) reported a slightly lower (11°C) minimum threshold temperature for loggerheads off NC. Coldest sea surface temperatures experienced on the middle continental shelf in the current study rarely declined below 17°C; however, warmest water temperatures regularly reached 29°C and did not elicit an avoidance response. The occurrence of loggerheads on the inner continental shelf in November also reflects historic distributional patterns. Ulrich (1978) reported that loggerheads were collected by trawling from inshore waters off SC in November 1977, and loggerheads were also trawl collected (albeit less frequently than in other months) from the Charleston, SC, channel in November and December 1991-1992 (Van Dolah and Maier, 1993; Dickerson et al., 1995).

Strong inverse correlations between transmitter temperature and dive duration and loggerhead location were observed. At the coldest water temperatures, loggerheads made less frequent but longer duration dives, as well as spent substantially more time at the sea surface overall. This pattern was repeated annually, though with varying degrees of magnitude. In addition to repetition within this study, this seasonal change in diving behavior for loggerheads has also been reported for loggerheads over-wintering on the continental shelf off of NC (Mansfield, 2006) and in the GOM (Renaud and Carpenter, 1994) as well upon encountering cool water upon return to inshore foraging areas in the spring (Keinath et al., 1995; Mansfield, 2006). Juvenile loggerheads over-wintering off the coast of Italy are also reported to exhibit many of the traits reported here. Specifically, juvenile loggerheads over-wintered relatively close (<80km) to where collected during warm water periods and adjusted dive behavior accordingly. Most winter dives lasted at least three hours (Hochscheid et al., 2007) and some dives lasted more than seven hours (Hochscheid et al., 2005). Additionally, several loggerheads moved among multiple locations off the Italian coast during the winter, a pattern which was distinctly observed for one transient loggerhead (ID#64550) in our study that actually 'rode' the Gulf Stream from Juno Beach, FL to Cape Hatteras, NC, before traversing back to northern FL during the winter. Movement on the continental shelf during winter was also noted (though to a lesser extent) for several resident loggerheads, particularly during brief warming periods.

Dive duration was only weakly associated with other environmental factors, of which wind speed was the closest match. Maier et al. (2004) suggested that loggerheads may relocate to deeper and presumably calmer waters at higher wind speeds and subsequent wave heights; however, principal components analysis only moderately supported this assertion. Although loggerheads may also seek refuge in shipping channels to avoid rougher seas (see Chapter 3) which would have kept their status as 'within the trawl boundaries', loggerheads located on the surrounding shoals infrequently returned to the Charleston, SC, shipping entrance channel. Perhaps less energy may be expended by simply 'hunkering down' to wait out a storm than by relocating to the channel or further offshore. Thus, although the suggestion of loggerhead movement to

deeper water during rough seas is plausible, reduced effectiveness of trawling gear to collect loggerheads at wind speeds >15kts may be a more appropriate explanation (Maier et al., 2004). Additionally, inability to detect a strong relationship between spatial distribution and wind speed in the current study was also inhibited by a lack of total observations of days with mean wind speeds >12 kts (9% of the 4,208 ‘turtle days’ evaluated). On days when mean wind speed exceeded 12kts, resident loggerheads were located within the trawl survey area, inshore and slightly offshore, as well as further offshore.

Observations from this study have far-reaching implications for the design of trawl surveys to estimate loggerhead relative abundance, as well as to determine their probability of detection. Seasonal occurrence of loggerheads on the inner continental shelf between April and November is highly predictable and strongly correlated with water temperature. Proportionately few transient loggerheads were monitored during this study, which may reflect collection of loggerheads no earlier than mid-May, which is four to six weeks after loggerheads first return inshore, and therefore most transients may have already moved through the area. Definitively outward (size or phenotypic qualities) or inward (genotype, gender) indications of predisposition for movement (large- or small-scale) could not be determined. Environmental predictors of fine-scale movement were also not elucidated, which may reflect the relatively small sample size ( $n=34$ ) of turtles monitored in this study and the high degree of individual variability among them. Consequently, the ability to accurately determine fine-scale loggerhead “detectability” in a particular location at a particular point in time would be incredibly difficult; thus, attempts to assess the probability of loggerhead occurrence may benefit the most from assessing large-scale (i.e., regional) changes in hydrographic influence, particularly in the context of density gradients (Epperly et al., 1995b). Therefore, random and extensive sampling, in conjunction with region-wide monitoring of environmental conditions at the time of sampling, may be the most appropriate way to assess the probability of loggerhead detection. Random and extensive sampling should also effectively combat habitat bias, given that loggerhead distributions are clustered (Chapter 1 and this chapter), and that clustered distributions may coincide with live-bottom habitats which occur as small, patchily distributed outcroppings (SEAMAP-SA, 2001). Furthermore, randomized sampling reduces the probability of localized damage to the seafloor, which could deter loggerheads from continued residence at what would have otherwise been a favorable habitat. Lastly, if clustered loggerhead distributions are actually associated with dense live-bottom habitats as suggested by Hopkins-Murphy et al. (2003), under-sampling of these habitat types (to avoid gear loss and habitat damage) would actually represent a conservative estimate of loggerhead relative abundance. Indeed, <10% of our regional trawling events since 2000 have occurred at stations classified as hard or probable hard bottom habitat (see Chapter 7).

**Table 4.1.** Demographics and distributional pattern of 34 juvenile loggerheads satellite-tagged following collection from the Charleston, SC, shipping entrance channel (2004-2007).

Tagged	TurtleID	Tag	SCLmin (cm)	Haplotype	Sex	Daily locations	Initial Disposition
Jun-04	CC0297	49120	60.6	CC-A01	F	310	resident
Jun-04	CC0296	49123	65.8	CC-A01	M	282	resident (left shelf)
Aug-04	CC0337	49121	60.5	CC-A01	F	263	resident
Aug-04	CC0329	49122	57.4	CC-A10	M	259	resident
Aug-04	CC0334	49124	57.9	CC-A01	M	172	resident
Aug-04	CC0330	52600	60.1	CC-A02	F	74	resident
May-05	CC0364	57683	57.7	CC-A02	F	136	resident
May-05	CC0360	57684	56.6	CC-A02	F	30	transient - Long Bay
May-05	CC0358	57685	72.9	CC-A01	F	293	resident
May-05	CC6045	57686	68.5	CC-A01	F	85	resident
May-05	CC0352	57687	62.3	CC-A01	F	268	resident (left shelf)
May-05	CC0349	57688	59.3	CC-A02	F	146	resident
Aug-05	CC0383	58939	62	CC-A02	M	68	resident
Aug-05	CC0381	58940	70	CC-A01	F	185	resident (left shelf?)
Aug-05	CC0378	58941	68.3	CC-A02	F	220	resident
Aug-05	CC0379	58942	59.8	CC-A02	F	233	resident
Aug-05	CC0382	58943	64.5	CC-A01	F	239	resident
May-06	CC0389	64549	~70cm	CC-A01	M	59	resident
May-06	CC0428	64550	67.4	CC-A01	F	383	transient - FL
May-06	CC0399	64551	70.2	CC-A02	F	31	resident
May-06	CC0426	64552	69.3	CC-A01	F	117	transient - Long Bay
May-06	CC0393	64553	59.6	CC-A01	F	35	resident
May-06	CC0394	64554	64.8	CC-A01	F	29	resident
May-07	CC0437	73113	58.5	CC-A02	F	89	resident
May-07	CC0436	73114	70.7	CC-A01	F	287	transient - VA/DE
May-07	CC0434	73115	62.2	CC-A02	M	67	resident
May-07	CC0433	73116	60.8	CC-A01	F	89	resident
May-07	CC0432	73117	70.4	CC-A01	F	122	resident
May-07	CC0435	73118	60.7	CC-A02	U	100	resident
Aug-07	CC0441	73119	60.8	CC-A01	M	65	resident
Aug-07	CC0408	73120	67.9	CC-A14	F	242	resident
Aug-07	CC0442	73121	76.8	CC-A01	F	134	transient - Long Bay
Aug-07	CC0438	73122	68.7	no sample	F	47	resident
Aug-07	CC0443	73123	69.7	CC-A01	F	199	transient - Long Bay

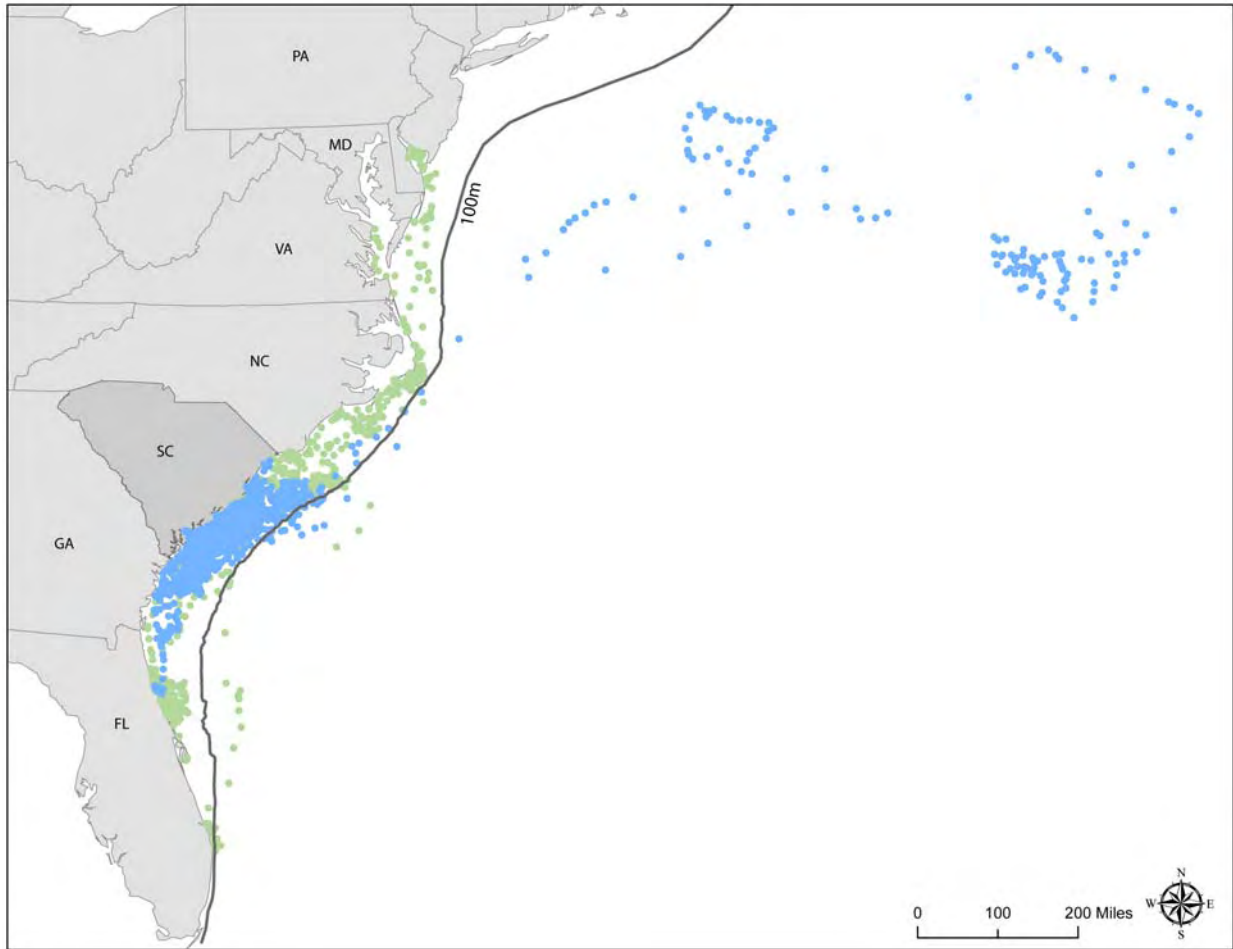
**Table 4.2.** Temporal distribution of daily location data collection for juvenile loggerheads.

Year	Feb-Apr	May-Jul	Aug-Oct	Nov-Jan	Total	% total
2004	n/a	81	352	421	854	16
2005	329	542	666	470	2007	37
2006	352	317	103	58	830	15
2007	80	437	667	271	1455	27
2008	199	13	n/a	n/a	212	4
Total	960	1390	1788	1220	5358	
% Total	18	26	33	23		

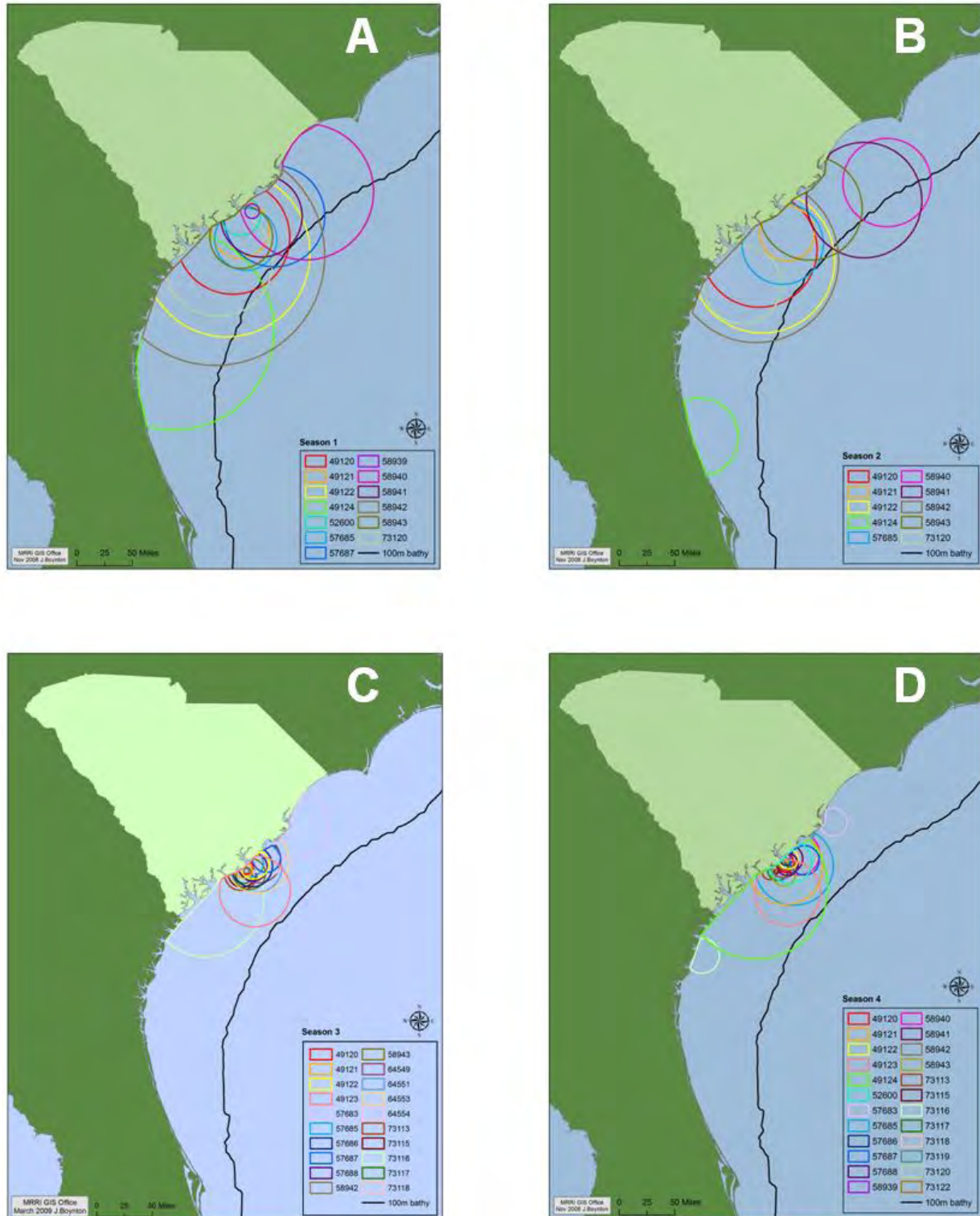
**Table 4.3.** Variance attributed to factors influencing the frequency of occurrence of loggerheads within the boundaries of the regional trawl survey area.

Metric	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigenvalue	4.127	3.0288	2.1658	1.5866	1.1231	0.9513	0.7755	0.67
Proportion	0.258	0.189	0.135	0.099	0.07	0.059	0.048	0.042
Cumulative	0.258	0.447	0.583	0.682	0.752	0.811	0.86	0.902

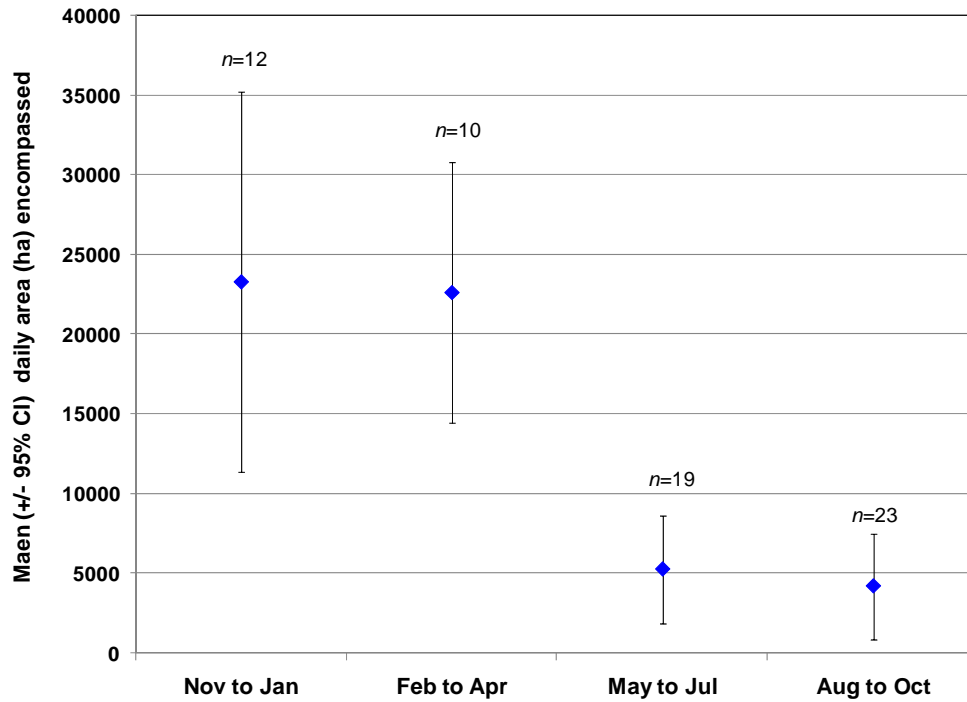
Metric	PC9	PC10	PC11	PC12	PC13	PC14	PC15	PC16
Eigenvalue	0.5081	0.3353	0.2202	0.1819	0.15	0.1115	0.0536	0.0113
Proportion	0.032	0.021	0.014	0.011	0.009	0.007	0.003	0.001
Cumulative	0.934	0.954	0.968	0.98	0.989	0.996	0.999	1



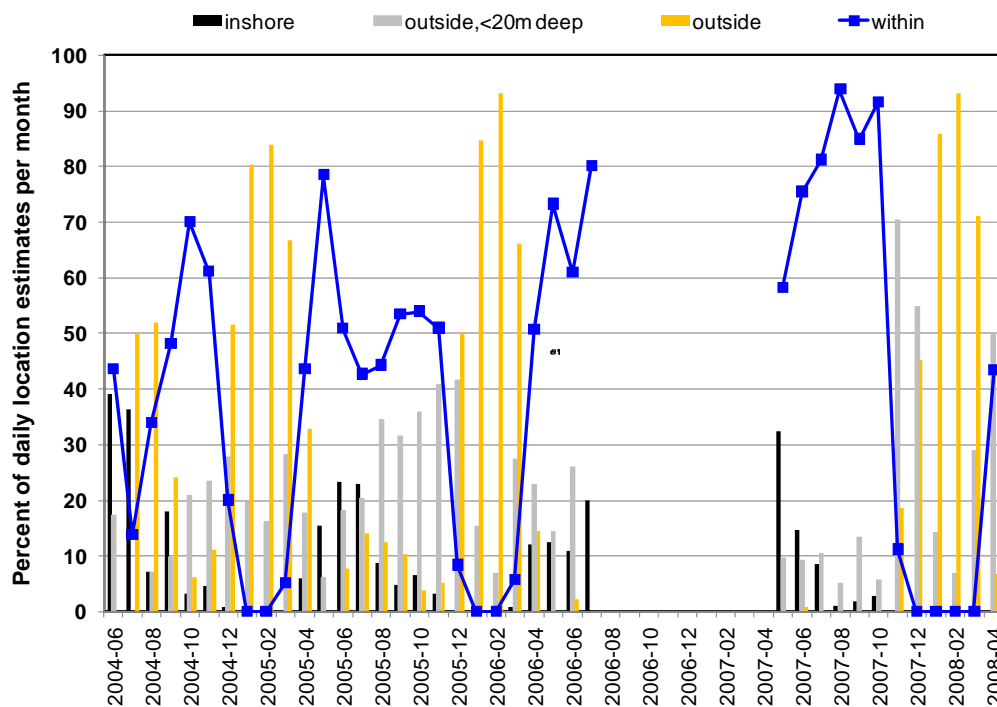
**Figure 4.1.** Geographic distribution of all daily location estimates for 28 resident (blue) and six transient (green) juvenile loggerheads satellite-tagged near Charleston, SC (2004-2007).



**Figure 4.2.** Seasonal centers of activity for 28 resident juvenile loggerheads. No difference in central location of activity centers was detected between November and January (A); February and April (B); May and July (C); or August and October (D).

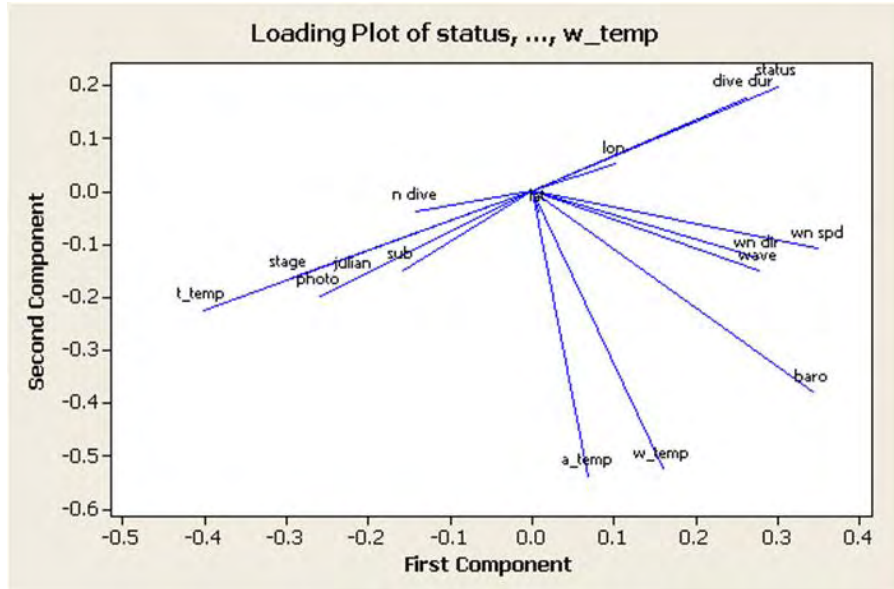


**Figure 4.3.** Seasonal variability in the area (continental shelf only) encompassed by resident loggerheads satellite-tagged between 2004 and 2007.

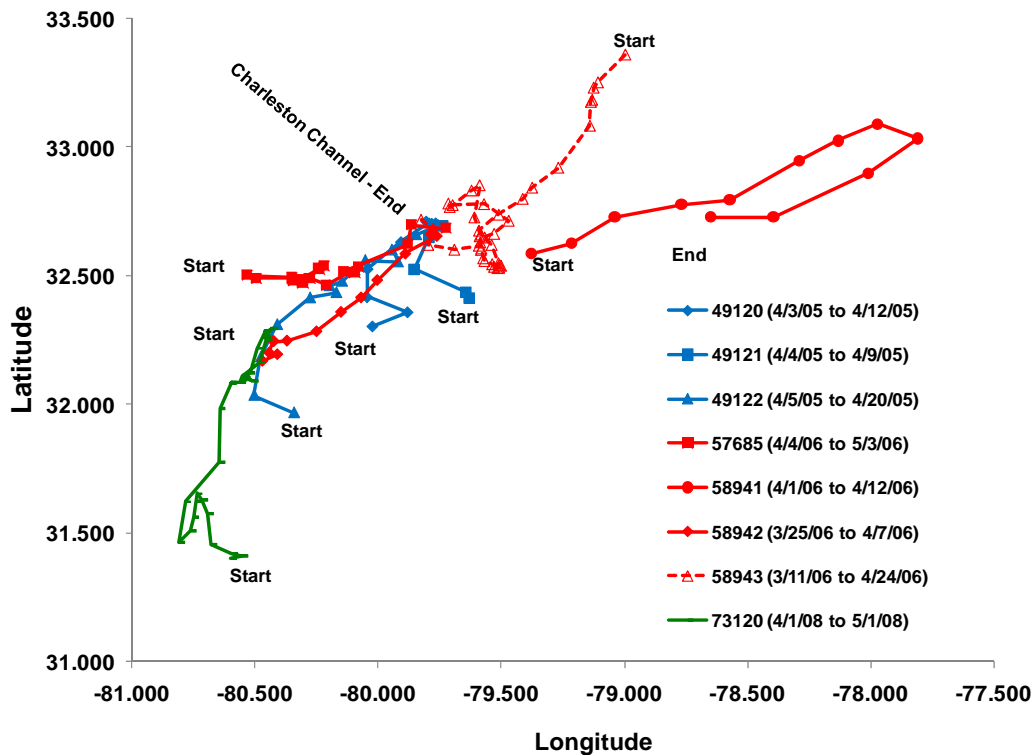


**Figure 4.4.** Seasonal variability in occurrence of resident loggerheads within the regional trawl survey area, 2004-2008.

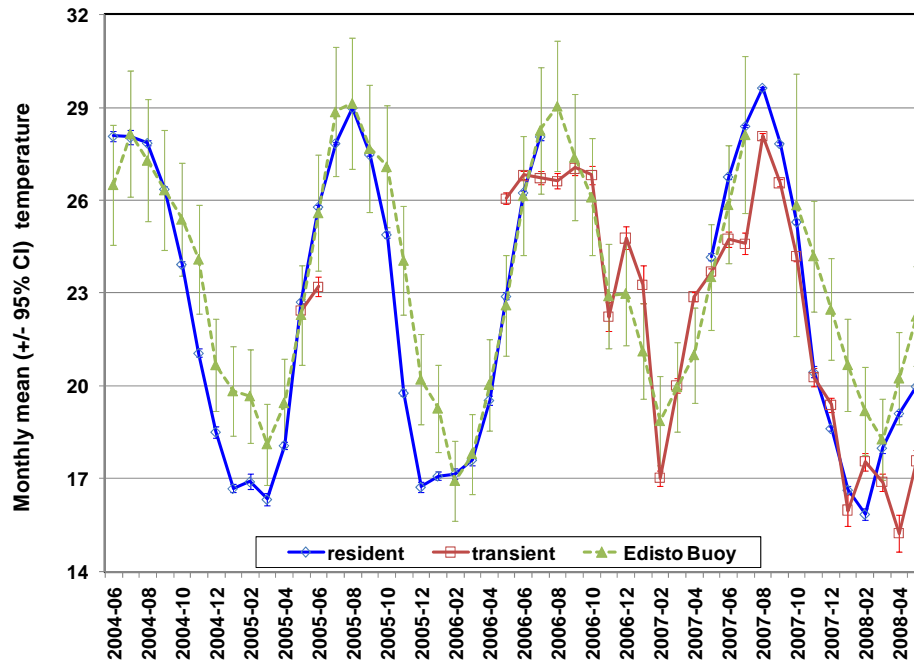




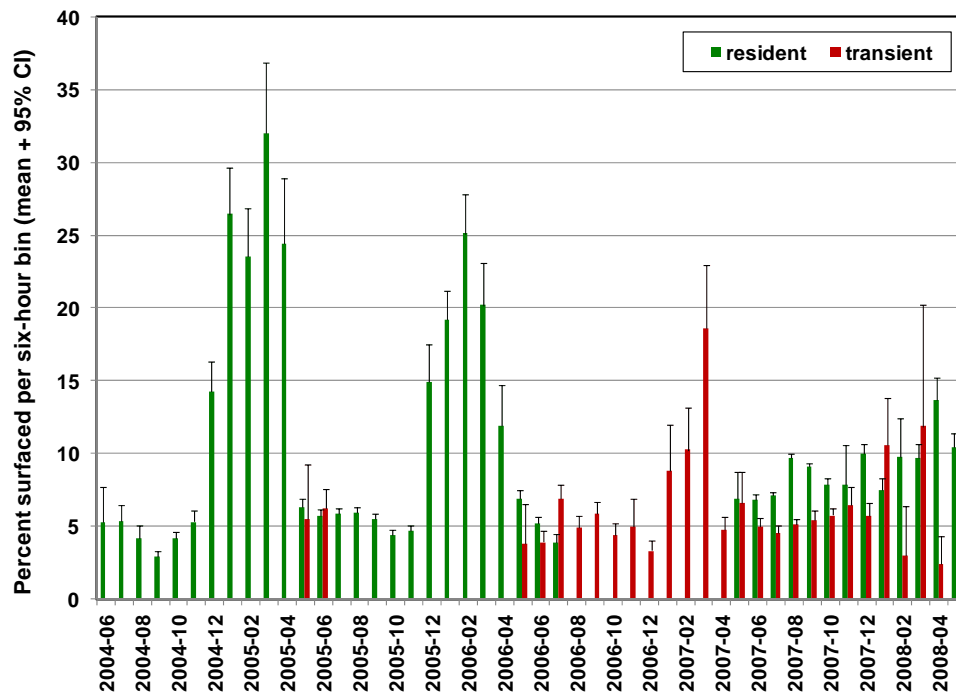
**Figure 4.5.** Correlation between the percent of loggerheads within the trawl survey boundary each day and eight temporal, hydrographic and meteorological factors, as determined by Principal Components Analysis.



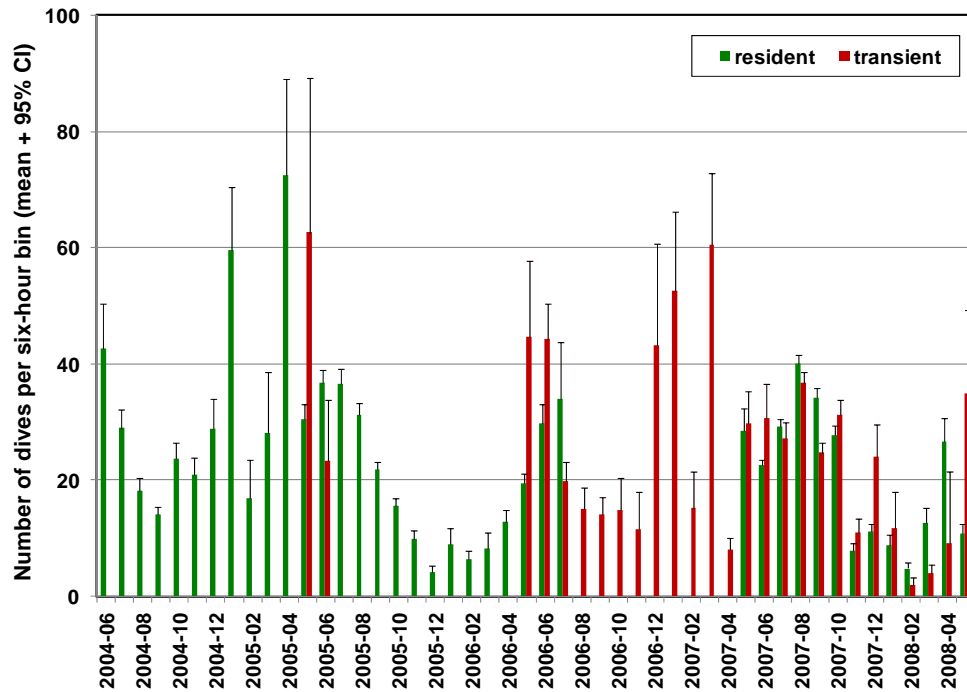
**Figure 4.6.** Trajectories for resident loggerheads returning to the vicinity of the Charleston, SC, shipping entrance channel after over-wintering in deeper waters on the outer continental shelf.



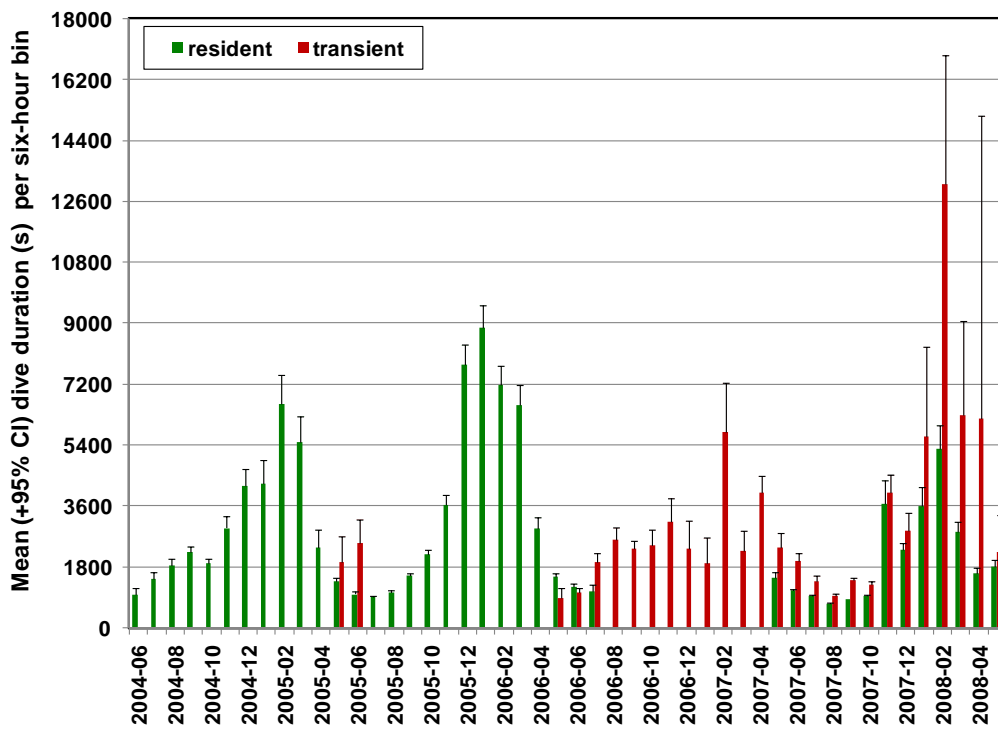
**Figure 4.7.** Monthly mean ( $\pm$  95% CI) transmitter temperatures for resident and transient loggerheads.



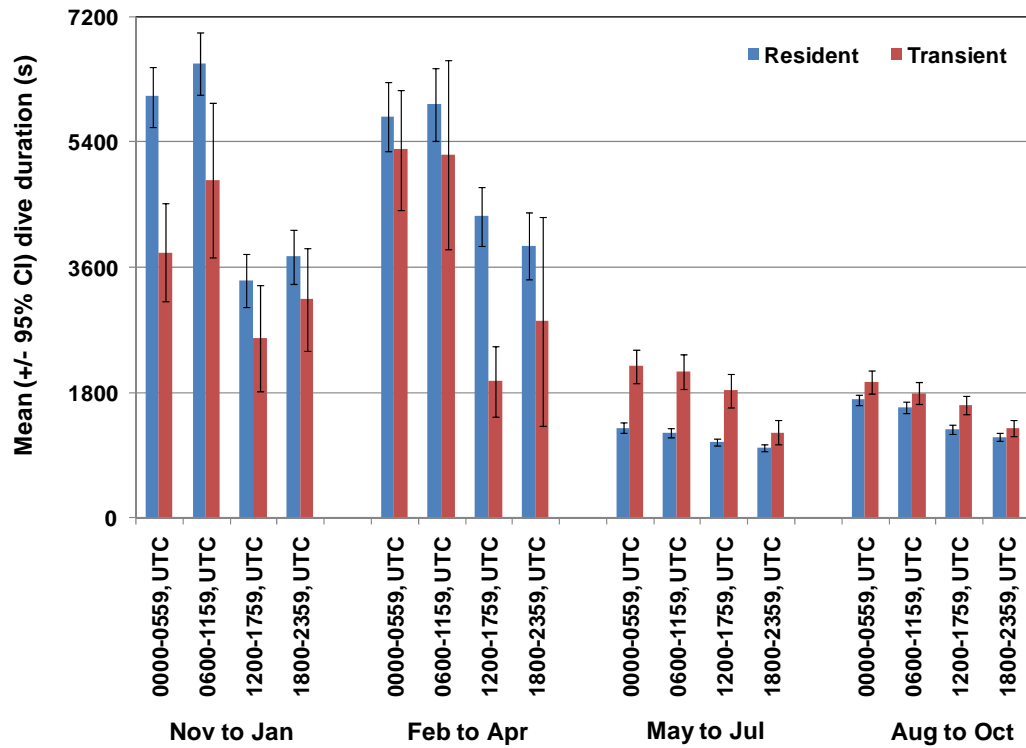
**Figure 4.8.** Seasonal variation in percent of time loggerheads remained at the surface during each six-hour data collection bin.



**Figure 4.9.** Seasonal variation in the number of dives made per six-hour data collection period.



**Figure 4.10.** Seasonal variation in mean dive duration (s) per six-hour data collection period.



**Figure 4.11.** Diel trends in mean dive duration (s) per six-hour data collection period.

## **Chapter 5 Seasonal distribution and activity of adult male loggerhead sea turtles.**

### **Introduction**

Excluding green turtles (Dizon and Balazs, 1982), adult male sea turtles rarely come ashore; thus, relatively less is known about their life history than is known for adult females and juveniles which present more opportunities for study. As more information regarding adult male sea turtles becomes available, the pivotal role they play in population dynamics becomes more apparent. Male-mediated gene flow is reported for sea turtles (Fitzsimmons et al., 1997; Bowen et al., 2005), and male-mediated dispersal (a precursor for colonization) has also been suggested (Casale et al., 2002). Sea turtle gender is determined by nest incubation temperature (a function of air temperature, precipitation and nest shading) with temperatures below 28°C typically producing  $\geq 90\%$  males (Standora and Spotila, 1985). Given projections for warming air temperatures in the coming decades, it has been suggested that adult male sea turtles may become less abundant as nest temperatures favor greater female production. In response to warmer air and water temperatures, annual sea turtle nesting may commence earlier (Hawkes et al., 2007), effectively preserving historic sex ratios. However, future nest temperatures in lower latitudes could also warm sufficiently to produce debilitating or lethal effects (Standora and Spotila, 1985); thus, offspring from areas with historically lower female production could become increasingly important, both with respect to producing viable hatchlings as well as regarding the simple production of males (Hawkes et al., 2007).

Throughout most of the Eastern Seaboard of the U.S., the extent of knowledge for adult male loggerheads has been derived from their infrequent collection in trawl surveys (Butler et al., 1987; Dickerson et al., 1995; Maier et al., 2004). Although these observations provide empirical data on temporal and spatial occurrence, limited biological data and/or small sample sizes hinder the conclusiveness of these data. In contrast, collection of at least 500 adult male loggerheads from the Port Canaveral, FL, shipping entrance channel since 1978 constitutes the epicenter of adult male loggerhead knowledge in the Western North Atlantic Ocean (Kemmerer et al., 1983; Butler et al., 1987; Henwood, 1987b; Nelson et al., 1987; Wibbels et al., 1987; Standora et al., 1993a,b, 1994; Dickerson et al., 1995; Blanvillain et al., 2008). Seasonal occurrence (Henwood, 1987b) and reproductive physiology (Wibbels et al., 1987; Blanvillain et al., 2008) have been extensively studied; however, considerably less emphasis has been placed on understanding habitat utilization and distributional patterns. Short-term distribution data for just six adult males (among 120 loggerheads tracked using radio and/or acoustic telemetry) are available for this location (Kemmerer et al., 1983; Nelson et al., 1987; Standora et al., 1993b). Though abundant in April and May, adult male loggerheads emigrate away from the channel and at least some portion of the population moves directly offshore (Hoffman and Fritts, 1982); however, because adult males tagged in Canaveral are rarely sighted elsewhere (Henwood, 1987b), information regarding non-resident males prior to the current study was historically non-existent. In order to address a critical data void, we initiated satellite-telemetry studies with adult male loggerheads beginning in 2006. In addition to reproductive physiology studies (Blanvillain et al., 2008), the objectives of this study were to characterize and describe the seasonal distribution patterns and diving behavior of adult males collected from a historically predictable spring-time aggregation (presumably for mating) near Port Canaveral, FL.

## Methods

### *Data collection and management*

Sea turtle and data collection are described in the General Methods section. Following standard data collection and assessment of reproductive condition (Blanvillain et al., 2008) satellite transmitters were attached in the same manner as described in Chapter 4. Transmitter data collected for adult male loggerheads satellite-tagged in 2006 was also identical to transmitter data collection described in Chapter 4. In 2007, five transmitters contained a dive depth counter, which provided the frequency distribution at selected depth (m) bins during six-hour data collection periods, as well as specific depth (m) during 10 equally-spaced points during the last dive prior to surfacing. Time at depth was analyzed for this report; however, due to lack of replicate observations for evaluating dive profiles, dive profile data were not analyzed. Depth bins selected for this study were as follows: 0-5m; 6-10m; 11-15m; 16-20m; 21-25m; 26-50m; 51-75m; 76-100m; and 101-127m. Due to data bit transmission constraints, depth-sensitive transmitters were not able to provide standard dive cycle metric data (n dives, mean dive duration and percent of time submerged); however, those data were available for the 15 non-depth-sensitive transmitters deployed in 2007.

Similar to juvenile loggerheads, data management relied on STAT (Coyne and Godley, 2005). Daily location estimates and standard transmitter sensor fields for adult male loggerheads were proofed and derived in the same manner as described for juvenile loggerheads in Chapter 4. Proofing of depth layer data made use of an additive feature, such that if the combination of data point values in each six-hour data collection bin exceeded “128” (maximum data bytes possible), all data points for the record were removed. Visual inspection of all records for congruency among repeated measurements was also conducted; discrepancies were reconciled where possible, otherwise all non-similar values were removed.

### *Data analysis*

Disposition status (resident versus transient) was assigned to adult male loggerheads monitored until at least 7 June of each year (date by which all transients and residents had moved away or offshore, respectively). If monitoring ceased before 7 June without a definitive movement to the north or south (transients) or offshore (residents), disposition status was listed as “unknown”.

Corollary data sets were obtained for analyzing spatial and behavioral data. Hourly sea surface temperature data were obtained from the National Data Buoy Center (NDBC) for three oceanographic buoys: Station #41009 located in 44m of water 37km east of Cape Canaveral, FL; Station #44014 located in 47m of water 119km east of Virginia Beach, VA; and Station #41014 located in 28m of water 48km southeast of Cape May, NJ.

Statistical testing was completed using Chi-square analyses in MS Excel, or using Kruskal-Wallis non-parametric analysis of variance by ranks in Minitab 15® (Minitab, Inc.). Chi-square tests were used to test for demographic differences among years and/or spatial disposition patterns. Rank testing was used to compare dive cycle metrics among groups and periods of time. Descriptive statistics were used to characterize diving activity with respect to water depth and sea surface temperature distributions.

## Results

### *General*

Twenty-nine adult male loggerheads were satellite-tagged between 2006 ( $n=9$ ) and 2007 ( $n=20$ ). Mean telemetry data collection was 144 days (range = 7 to 366). No adult males satellite tagged in 2006 were detected after December 2006; however, over-wintering (February to March 2008) data were collected for three adult males tagged in 2007 (Table 5.1).

Fifty-five percent ( $n=16$ ) of loggerheads were transients, 38% ( $n=11$ ) were resident and disposition could not be assessed for two (7%) loggerheads. Fourteen percent ( $n=4$ ) of adult male loggerheads were reproductively inactive (Blanvillain et al., 2008), and all inactive males remained resident; thus, a significant difference (Chi-square<sub>df=4,  $\alpha=0.05$</sub> ,  $X=7.59$ ,  $p=0.022$ ) in the proportion of inactive loggerheads among resident and transient loggerheads was detected.

Mean size of satellite-tagged loggerheads was 92.8cm SCLmin (range = 86.6 to 107.0cm). No statistical differences in loggerhead size was detected among years (K-W,  $df=1$ ,  $p=0.741$ ) or between two collection periods in April 2007 (K-W,  $df=1$ ,  $p=0.545$ ). A significant size difference (K-W,  $df=1$ ,  $p=0.026$ ) was detected among dispositions, with larger median size observed for transient (95.2cm SCLmin) compared to resident (88.9cm SCLmin) loggerheads.

Genetics data were available for 28 of 29 satellite-tagged adult male loggerheads. Fifty-seven percent ( $n=16$ ) of loggerheads were of the CC-A01 haplotype, 37% ( $n=10$ ) loggerheads were of the CC-A02 haplotype, and CC-A04 and CC-A20 were each identified once. No statistical differences were noted for haplotype distributions among years (K-W,  $df=2$ ,  $p=0.539$ ) or among transient versus resident dispositions (K-W,  $df=2$ ,  $p=0.055$ ).

Temporal collection data across years suggested a progressive increase in the proportion of transient adult males throughout the month of April (Figure 5.1); however, this trend was not statistically significant (K-W,  $df=4$ ,  $p=0.391$ ).

### *Spatial distribution*

A total of 2,975 daily location estimates for 29 adult male loggerheads were analyzed. Forty-five percent of daily location estimates ( $n=1,333$ ) were obtained during an 11-week period spanning 1 April through 16 June, with 57% ( $n=762$ ) of daily location estimates obtained for transients, 39% ( $n=565$ ) for residents, and 4% ( $n=50$ ) for loggerheads of unknown distribution. A distinct distribution shift in mid-May (Figure 5.2) possibly signaled cessation of mating.

Between 1 April and 5 May, resident and transient adult male loggerheads were well-mixed in near shore waters, generally within 20km of shore (Figure 5.3a,b). Transients moved further offshore and started to emigrate away from Canaveral between 6-12 May; however, the emigration of transient males was most evident between 13-26 May (Figure 5.3c). Movement of resident males offshore of -80.5°W longitude became apparent after 20 May (Figure 5.3c). After 3 June, no spatial overlap existed between transient and resident males (Figure 5.3d). Between June and November (the extent of monitoring for residents), all daily locations estimates for resident males occurred between -79.8°W and -80.8°W longitude and 27.6°N to 29.2°N latitude.

Transient adult male loggerheads relocated to numerous destinations in the Western Atlantic and Gulf of Mexico where they remained highly localized (Figure 5.4). Twice as many transient males traveled north ( $n=11$ ) as went south ( $n=5$ ). Four adult males settled in near shore waters between SC and NC, four settled in a variety of water depths off the coasts of VA and MD, and two occupied mid-shelf waters off NJ. Destination for one north-bound male could not be determined. South-bound males took up residence in shallow waters associated with the FL Keys ( $n=2$ ), the FL panhandle ( $n=2$ ) and the Bahamas ( $n=1$ ). With the exception of transient males in the FL Keys, off VA and off NJ, transient males became established at locations 100-250km apart from the next closest known transient male location. Over-wintering distribution was documented for three transient adult males; two remained in the northern Gulf of Mexico, and an adult male off VA relocated south to the shelf-slope break ESE of Cape Fear, NC.

#### *Temperature distribution*

Ninety-five percent of transmitter temperature records ( $n=27,223$  of 28,804) were analyzed. Seasonal disparity in the reliability of temperature records was noted, with data retention rates <75% during the last months of data collection (December 2006 and February-March 2008).

In April 2006 and 2007, mean transmitter temperature for resident males closely resembled sea surface temperature (SST) on the middle continental shelf east of Cape Canaveral; however, between May and August of both years, mean transmitter temperature was substantially cooler (by 1-3°C) than sea surface temperature (Figure 5.5). Pronounced increase in mean transmitter temperature for resident males occurred during September in both years, and remained within 1°C of SST for the duration of monitoring throughout the fall.

Transient males inhabited a variety of thermal regimes (Figure 5.6). Males located between SC and southward experienced peak summer temperatures near or above 30°C, whereas males located between VA and NJ remained in waters 21-24°C during the summer. Mean transmitter temperatures for three males that resided north of Cape Hatteras, NC, and remained 50-100km offshore were substantially cooler (by 2-4°C) than SST during the summer (Figure 5.7).

#### *Diving Behavior*

Diving behavior was distinctly different between resident and transient loggerheads. With the exception of a transient loggerhead in SC during July 2007, resident loggerheads made fewer (Figure 5.8) and longer (Figure 5.9) dives than transient loggerheads during every month in which resident and transient data were collected. Resident loggerheads averaged dive durations <1800s (30min) in July and August 2006. Conversely, transient loggerheads only rarely averaged dive durations greater than 900s (15 min), with most dives  $\geq 900$ s occurring during April and May 2006 and 2007. Adult male loggerheads generally spent between 5-10% of the time at the surface; except for April 2007, when more than 10% of time spent at the surface was associated with colder water temperatures (Figure 5.10).

Dive duration was statistically different (K-W,  $df=3$ ,  $p<0.001$ ) with respect to time of day for both resident and transient (all groups) loggerheads. With the exception of transient loggerheads in the FL Keys and Bahamas between June and October, mean dive duration was always greater between 0000 and 1159 UTC than between 1200 and 2359 UTC (Figure 5.11).



### *Depth distribution*

Seventy-five percent of depth distribution records (n=4,690 of 6,244) representing 2,054 six-hour data collection bins were analyzed. Fifty-nine percent of usable depth data were collected during April and May 2007.

Pronounced shifts in depth distribution after moving away from Cape Canaveral, FL, were noted for five male loggerheads; however, considerable variability was observed (Figures 5.12 to 5.16). During April and May, loggerheads generally remained in water depths 6-15m deep.

Three male loggerheads briefly occupied waters  $\geq 21$  m before moving offshore or emigrating away from Cape Canaveral, FL. On 9 May, ID#73095 relocated to the Northeast of Cape Canaveral and settled in an area  $>26$ m deep, where it remained at depth for 90-95% of data collection bins until contact was lost on 21 May (Figure 14). During April, ID#73097 was predominantly located in waters 11-15m deep, though also spent approximately 5% of each data collection bin in water 16-20m deep (Figure 16). On 20 May, an abrupt shift in depth distribution for this loggerhead occurred, which may indicate transmitter malfunction and/or switch to pelagic foraging. A transient male (ID#73094, Figure 13) nearly exclusively occupied waters 16-25m deep on 13 May and 23-24 May.

During emigration away from Canaveral, transient males primarily remained within 5m of the surface. While off the NC coast in early June, ID#73094 dove as deep as 75m; however, from mid-June through the duration of tracking, this loggerhead remained almost exclusively in water depths  $\leq 10$ m. ID#73093 rapidly became established in the FL Keys after leaving Canaveral, and exclusively remained in water shallower than 5m (Figure 12); data collected beginning in the middle of August may indicate pressure sensor failure and/or floating on the surface. Between July and October, ID#73096 averaged 25% of each six-hour period at the surface, 35% of each bin between the surface and 5m depth, 30% of each bin between 26-50m, and the remainder of time was generally spent transitioning between 50m and the surface. Between November and March, this male spent 70-90% of each collection bin in waters 26-50m deep with most of the remainder of time spent at the surface.

### **Discussion**

Data on adult male loggerheads acquired in this study yielded a wealth of distributional and behavioral information as these animals were collected from a large mating aggregation area adjacent to the second largest loggerhead rookery in the world (NMFS and USFWS, 2008). Most adult males were monitored during the mating season and until at least mid-summer, and nearly one-third of them were also monitored into the fall. The shortest duration track of six days (April 2006) exceeds the extent of tracking data for adult males using radio and acoustic telemetry previously available at this same location. Standora et al. (1993b) tagged three adult male loggerheads with depth-sensitive transmitters in spring 1993 and continuously monitored them for 20-48h after release; two males were regularly detected within a 3.5km radius of the shipping channel. Kemmerer et al. (1983) tagged and released three adult males with radio and acoustic transmitters near Cape Canaveral, FL, in fall 1981; no data were collected for one male and observation of the other two males (within 10km of the shoreline) was made over a 1-2d period, with no other information or description of movement provided.

Dispersal of adult male loggerheads away from Canaveral was highly synchronized; however, excluding Julian date, exact departure cues are uncertain. Resident males shifted to deeper waters offshore concurrent with rapid emigration of transient males away from Canaveral to distant and diverse destinations, some requiring long migrations to the Mid Atlantic Bight and northern Gulf of Mexico.

The geographically distinct Cape Hatteras, NC, may represent a seasonally important landmark for adult male loggerheads. One of two male loggerheads tracked to coastal waters off NJ in 2006 returned to the vicinity of Cape Hatteras, NC, in September, where it remained in close to shore for nearly a month before the transmitter ceased signal transmission. A second transient male in our study also returned to Cape Hatteras, NC, in November, before over-wintering near the edge of the continental shelf to the east/southeast of Cape Fear, NC. Similar findings have also been found for adult male loggerheads tracked by other researchers. In spring 2007, two adult male loggerheads were transferred to the SC Aquarium after entanglement in crab pot lines encountered in SC estuarine waterways. Subsequent satellite tracking of these two adult males following rehabilitation also revealed a northward transit to Cape Hatteras, NC, during the fall before over-wintering in deep water east/southeast of Cape Fear, NC (SCA & SCDNR, unpublished data). And two adult male loggerheads satellite-tagged in Chesapeake Bay in fall 1991 over-wintered in the same general vicinity off NC; these turtles occurred off of Cape Hatteras, NC, in late May, and re-entered the Chesapeake Bay in June (Keinath, 1993).

Transient males that established themselves between Cape Hatteras, NC, and major nesting beaches to the south, are strategically positioned to encounter (for eventual mating) adult female loggerheads on foraging grounds at over-wintering areas off NC (SCDNR, unpublished data 1). Although it is not known how receptive (if at all) post-nesting females would be to mating in the fall, or if males could even produce sperm in the fall (Wibbels et al., 1990), it is worth noting that sperm can be viably stored in reptiles (Gist and Congdon, 1998). Furthermore, although continued mating within the same season is unlikely, annual mating capability for adult males (Wibbels et al., 1990) suggests that male-mediated gene flow may also stem from physical relocation of males among breeding areas in successive years. Thus, although the occurrence of adult male loggerheads off NC during fall and winter may be completely unrelated to reproductive behavior, aggregation of both adult male and female loggerheads may present reproductive opportunities the following spring.

Approximately half of adult male loggerheads remained resident east of Cape Canaveral, FL, which offers a different set of reproductive and evolutionary advantages. Shorter migration distances conserve energy which can instead be used for building mass and (seasonally) reproductive output. Residence east of Canaveral may also allow adult male loggerheads to mate sooner than transient males, which may arrive later (and en masse) due to longer travel distances following the onset of environmental migratory cues such as warming water temperature and increasing photoperiod. Trawl surveys have repeatedly documented increasing occurrence of adult males in the Port Canaveral, FL, shipping entrance channel in March and peak occurrence in April (Henwood, 1987b; Dickerson et al., 1995). Wibbels et al. (1987) showed peak testosterone levels (albeit only measured for two adult males) in March, with slightly lower (mean = 145,000 pg/ml) values noted for 55 adult males collected in April. Although Blanvillain

et al. (2008) showed that low testosterone among adult males from Canaveral does not necessarily represent reproductively inactive turtles high testosterone levels among adult males collected in March and April by Wibbels et al. (1987) does indicate reproductive activity. Furthermore, one resident male which was recaptured in the Port Canaveral, FL, shipping entrance channel was reproductively active in both years of our study. Henwood (1987b) also suggested the possibility of annual mating capabilities for adult male loggerheads at this location. Thus, assuming that annual reproductive activity is representative, residence (and therefore being 'first in line') near Canaveral may represent a reproductive advantage given polyandry which has been documented for sea turtles (Fitzsimmons, 1998; Crim et al., 2002).

Resident males in our study were significantly smaller than transient males. Although size discrepancies between the two groups may be confounded by small overall sample sizes, a growing body of evidence suggests the possibility of size-related partitioning of foraging habitat among adult loggerheads, which warrants further discussion. Using a combination of stable isotope and satellite telemetry data, Hatase et al. (2002a) determined that three large adult female loggerheads shifted to neritic foraging grounds after the nesting season while two smaller females moved offshore and occupied oceanic habitats. In our study, many (though not all) transients also exhibited preference for relatively shallow, inshore waters as opposed to deeper waters on the middle to outer continental shelf. Furthermore, two smaller adult male loggerheads (78.3 and 78.4cm SCL) have also been tracked by satellite telemetry off the coast of Japan, both of which utilized the Kuroshio Current and occupied oceanic habitats (Sakamoto et al., 1997; Hatase et al., 2002b). However, it should also be noted that both adult male loggerheads were initially collected from inshore set-net fishing gear; thus, smaller adult males do not exclusively reside in oceanic habitats. Indeed, one of the adult males was tracked during the reproductive season (Sakamoto et al., 1997) and spatial distribution data suggest it was reproductively active, while the second adult male (Hatase et al., 2002b) was tracked during the winter and only for 35 days, so reproductive inferences were not possible.

The post-mating spatial distribution of adult male loggerheads sheds considerable light on their historical paucity in coastal trawl surveys within the SE USA. Transient males generally utilized the upper 5m of the water column during emigration, and predominantly transited via the middle to outer continental shelf. A notable exception was the passage of ID#64548 to the near shore waters off Charleston, SC, in late May 2006 en route to NJ. Late spring collection of adult male loggerheads from shipping channels along the GA (Nelson, 1996) and SC (Van Dolah and Maier, 1993; Dickerson et al., 1995; Chapter 3 of this report) coasts has also been reported, and may at least occasionally represent ephemeral channel utilization en route to other destinations. As elucidated in this study, other destinations include a number of spatially diverse coastal and estuarine waters (including residentially developed lagoons) which are too shallow to be sampled by 75' trawlers. Similarly, approximately half (predominantly residents) of adult male loggerheads satellite-tagged in this study relocated to waters 25-50m water depth, which are considerably deeper than water depths sampled by the commercial shrimp fleet and research trawl surveys, but potentially shallower than where fisheries observer efforts may be concentrated to monitor long-line fishing (NMFS, 2008). However, aerial surveys historically document loggerheads between summer and fall on the middle to outer continental east of Cape Canaveral, FL, near the western wall of the Gulf Stream (Schroeder and Thompson, 1987).

A variety of diving behaviors have been reported for loggerhead sea turtles. Yano and Tanaka (1991) describe six dive types, of which remaining at a constant depth is the most common. Diving behavior is influenced by time of day, circadian rhythm, tidal flow and sea state (Sakamoto et al., 1993; Sakamoto et al., 1990a,b). Diving patterns also differ between neritic and oceanic habitats (Polovina et al., 2000, 2003; Sakamoto et al. 1990a), with dives deeper than 200m (the generally accepted depth for the continental shelf-slope break) occurring for both juvenile and adult loggerheads in oceanic habitats. Circadian rhythm exerts the strongest influence on loggerhead diving behavior in oceanic environments, whereas tidal cycles exert greater influences on loggerhead diving behavior in neritic habitats (Sakamoto et al., 1990b).

Resident and transient adult male loggerheads generally made longer dives during the mating season than during the summer and early fall. Dive durations were distinctly different for transient and resident males during the mating season. During 2006, transients generally remained submerged for 15-20min while residents remained submerged for approximately three times as long. A similar relative pattern was repeated in 2007; however, dive duration for both residents and transients was approximately double dive durations observed the previous year. Although mounted pairs are regularly observed at the surface which has led to the supposition that mating occurs at the surface, neither submergence data nor limited depth distribution data suggested frequent utilization of surface waters during the mating season. Indeed, dive duration data suggest that mating more likely occurs while submerged, and that observations of mounted pairs at the surface may only occur when one or both turtles needs to return to the surface to breathe. Mounted loggerhead pairs have been observed at depth in Greek waters during April, with mounted pairs more commonly occurring under higher sea states and lower visibility, which likely enhances the effectiveness of surprising a potential mate (Schofield et al., 2006). Thus, a variety of logistical constraints have likely contributed to less frequent citing of mating at depth.

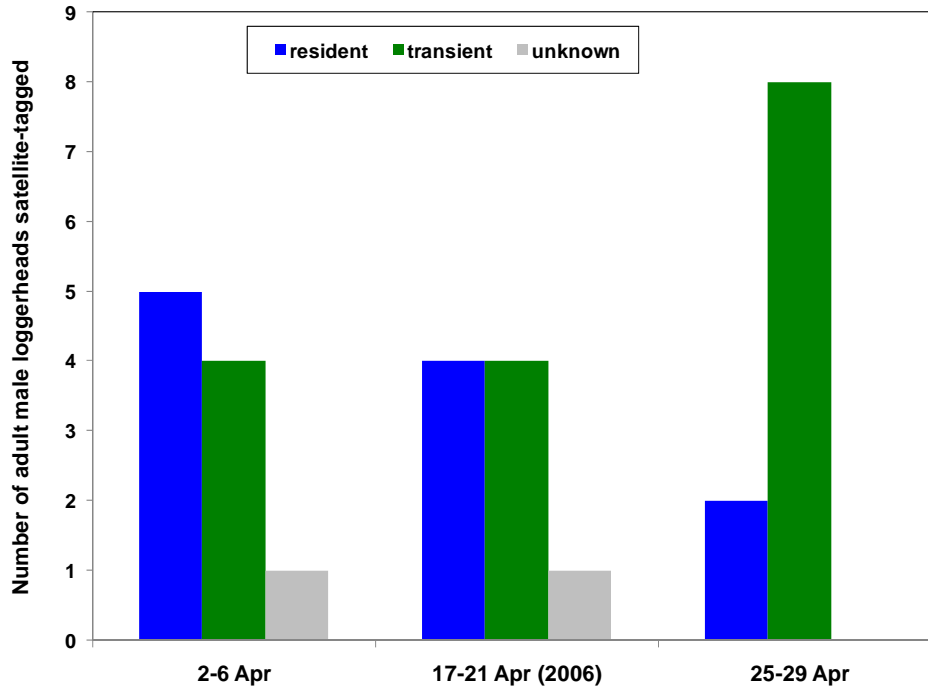
Diving behavior of adult male loggerheads in water depths >25m was characterized by frequent, short duration (<15min) dives. The pattern of both resident and transient adult male loggerheads utilizing water masses cooler than sea surface temperature between June and August was documented in subsequent years; thus, lending support to the validity of these observations. Depth distribution data for two transient males on the middle to outer continental shelf off VA substantiate that loggerheads briefly dive to the bottom at these depths; however, the majority of time was spent in the upper 5m of the water column. Thus, unless benthic prey items were abundant and could easily be located and consumed during short dives, depth distribution data suggest that adult male loggerheads may have been foraging on gelatinous zooplankton instead. Short duration and frequent dives below thermocline depth coupled with ingesting cold seawater to regulate body temperature is reported for adult female loggerheads in pelagic environments off Japan (Sakamoto et al., 1990). Seawater ingestion may also occur while foraging on salps and other gelatinous zooplankton which may congregate near thermoclines (Graham et al., 2001). Preliminary stable isotope results for adult male loggerheads tracked for this study do indeed suggest foraging on organisms inhabiting the water column rather than the sea floor (Reich, pers. comm.), which may in turn affect the susceptibility adult male loggerheads to collection, both for directed surveys and as fisheries by-catch.

**Table 5.1.** Demographic and distribution data for 29 adult male loggerheads satellite-tagged after collection from the Port Canaveral, FL, shipping entrance channel in April 2006 and 2007.

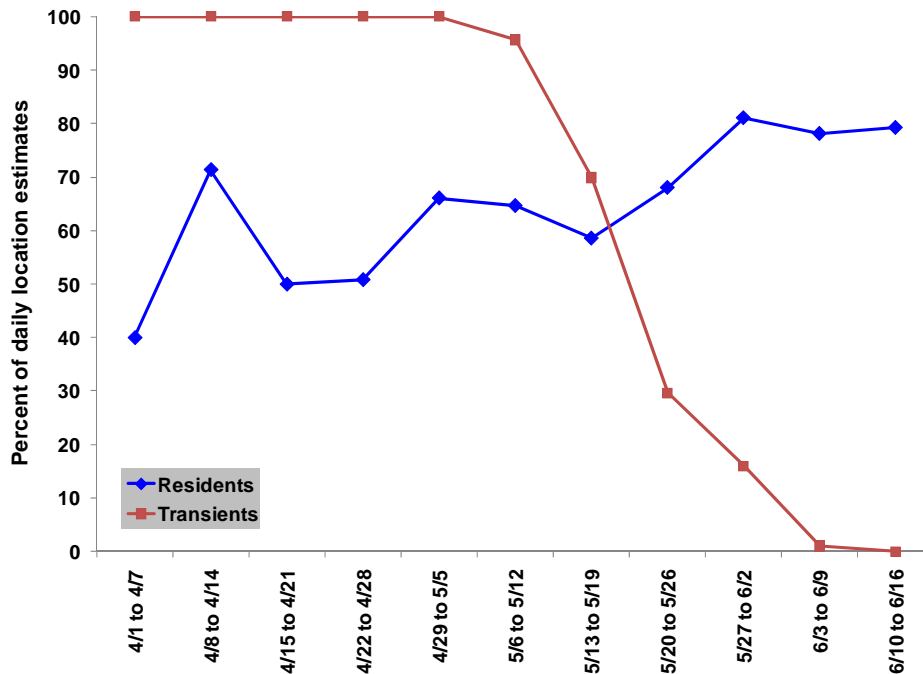
Tag	SCLmin (cm)	Haplotype	Reproductive Activity*	Disposition	Begin	End	Days
64540	86.6	CC-A01	active	resident	19-Apr-06	06-Jul-06	79
64541	>95	CC-A01	active	unknown	18-Apr-06	24-Apr-06	7
64542	87.2	CC-A02	not active	resident	18-Apr-06	24-Nov-06	221
64543	89.5	CC-A01	active	transient - SC	17-Apr-06	21-Jul-06	96
64544	>95	CC-A02	active	transient - MD	18-Apr-06	19-Aug-06	124
64545	90.7	CC-A02	not active	resident	19-Apr-06	16-Dec-06	242
64546	88.9	CC-A01	not active	resident	19-Apr-06	25-Jun-06	68
64547	97.4	CC-A02	active	transient - NJ	20-Apr-06	16-Aug-06	119
64548	91.6	CC-A01	active	transient - NJ	19-Apr-06	30-Sep-06	165
73093^	86.9	CC-A01	active	transient - FL Keys	26-Apr-07	27-Dec-07	246
73094^	102.5	CC-A01	active	transient - VA	28-Apr-07	16-Aug-07	111
73095^	88.7	CC-A02	not active	resident	03-Apr-07	21-Jun-07	80
73096^	101	CC-A01	active	transient - VA	29-Apr-07	28-Apr-08	366
73097^	90.3	CC-A20	active	resident	02-Apr-07	15-Jul-07	105
73098	89	CC-A01	active	transient - NC	27-Apr-07	20-Sep-07	147
73099	102	CC-A02	active	transient - Bahamas	28-Apr-07	02-Sep-07	128
73100	102.8	CC-A02	active	transient - NC	04-Apr-07	05-Aug-07	124
73101	89.5	CC-A01	active	transient - FL panhandle	26-Apr-07	22-Feb-08	303
73102	86.8	CC-A01	active	transient - north, unknown	03-Apr-07	05-Jun-07	64
73103	87.2	CC-A01	active	resident	04-Apr-07	06-Jul-07	94
73104	89.6	No sample	active	resident	26-Apr-07	26-Oct-07	184
73105	87.4	CC-A01	active	transient - SC	28-Apr-07	24-Oct-07	180
73106	102.1	CC-A01	active	transient - VA	29-Apr-07	17-Aug-07	111
73107	87.9	CC-A01	active	unknown	04-Apr-07	21-May-07	48
73108	87.3	CC-A02	active	resident	06-Apr-07	17-Aug-07	134
73109	93.2	CC-A02	active	resident	27-Apr-07	24-Aug-07	120
73110	93.4	CC-A04	active	transient - FL Keys	06-Apr-07	05-Aug-07	122
73111	107	CC-A01	active	transient - FL panhandle	06-Apr-07	26-Mar-08	356
73112	91.1	CC-A02	active	resident	05-Apr-07	07-May-07	33

\*Determined by various methods, including testicular biopsy (Blanvillain et al., 2008)

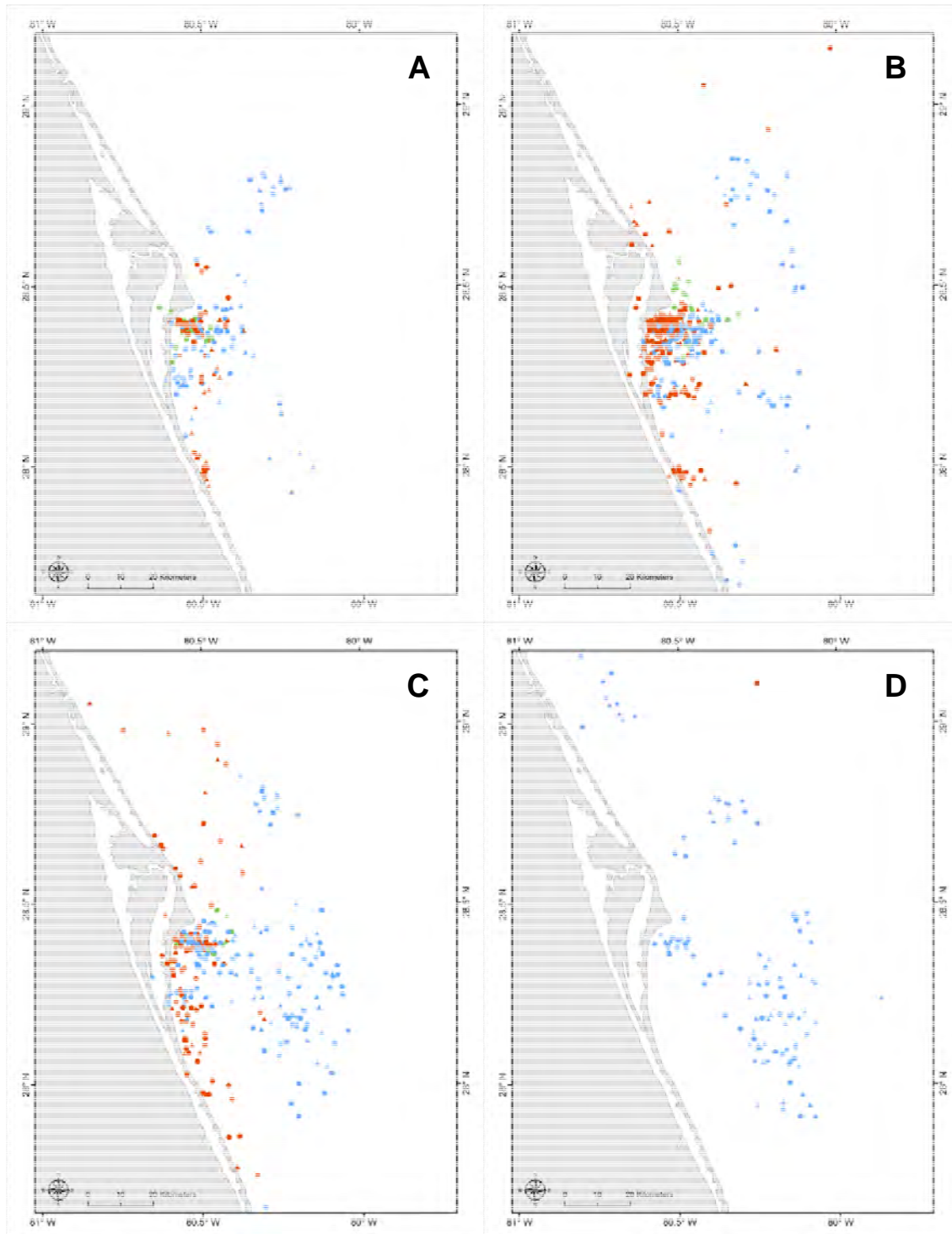
^Depth-sensitive transmitter



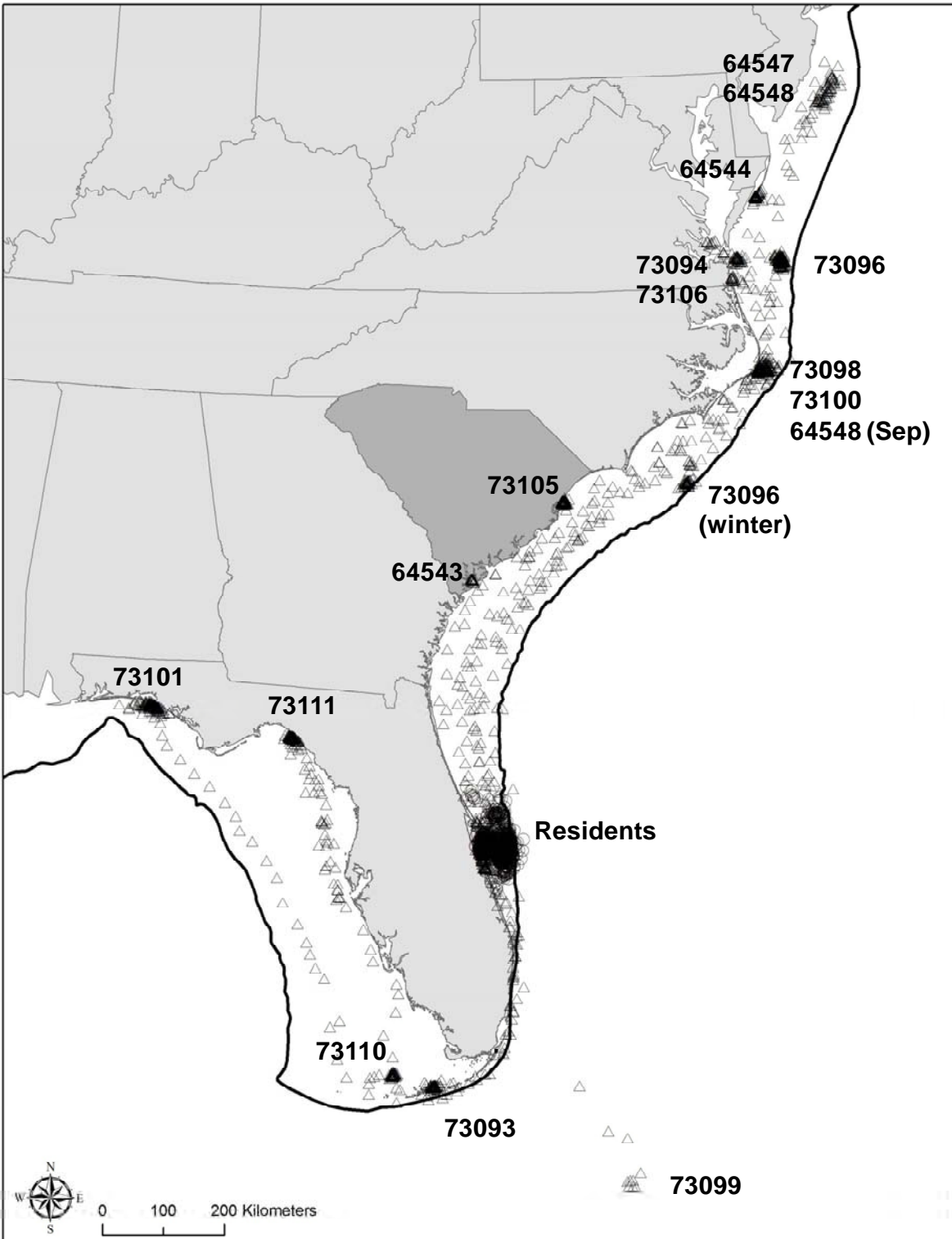
**Figure 5.1.** Temporal distribution of adult male loggerheads collected from the Port Canaveral, FL, shipping channel (2006-2007).



**Figure 5.2.** Temporal shift in daily location estimates for resident (% offshore of  $-80.5^{\circ}\text{W}$ ) and transient (% within area occupied by residents) adult male loggerheads located near Cape Canaveral, FL, between April and mid-June.

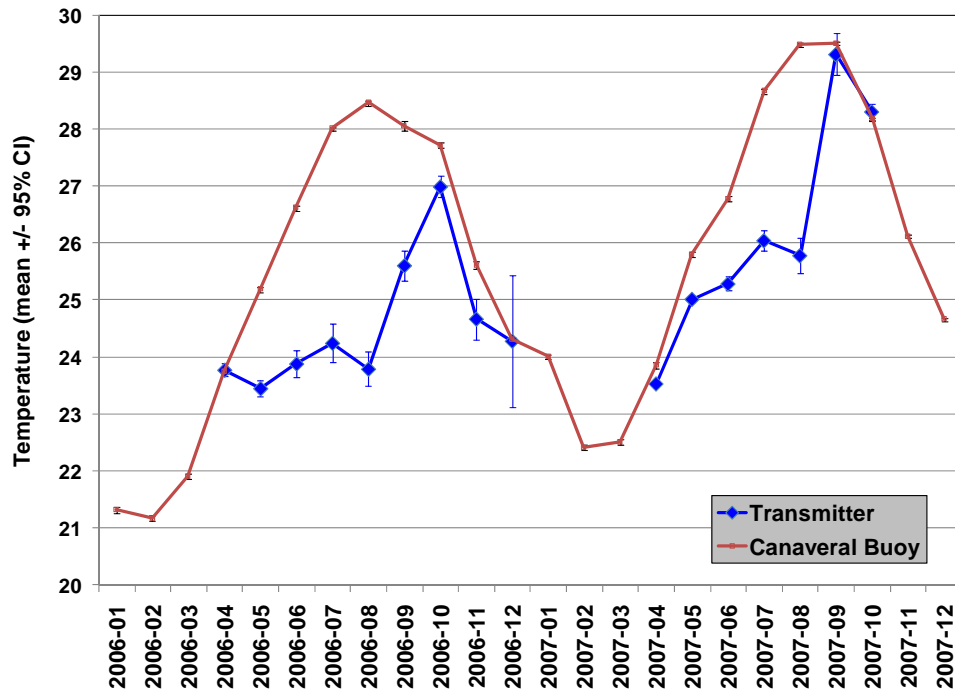


**Figure 5.3.** Spatial distribution of resident (blue), transient (orange), and unknown (green) distribution adult male loggerheads near Cape Canaveral, FL, between April and mid-June. Symbols denote consecutive seven-day observation periods (circle = first; triangle = second; square = third) between 1 and 21 April (A), 22 April and 12 May (B), 13 May and 2 June (C), and 3 to 16 June (D).

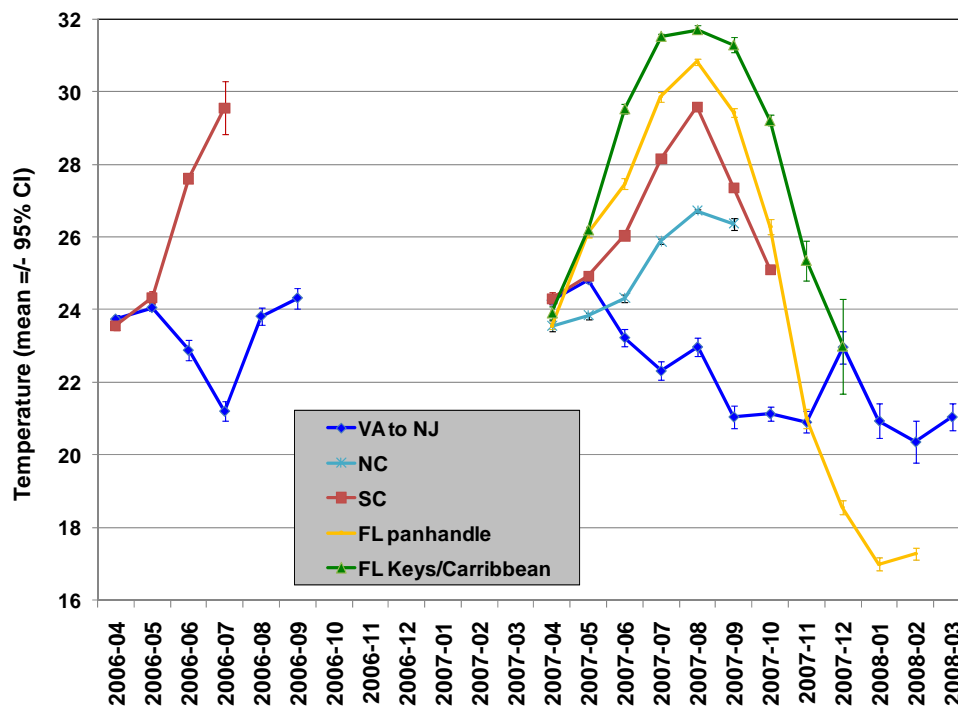


**Figure 5.4.** Spatial distribution patterns of resident (circles) and transient (triangles) adult male loggerheads after collection and satellite-tagging from the Port Canaveral, FL, shipping entrance channel in April 2006 and 2007. Dark areas indicate repeated detection for a particular location.

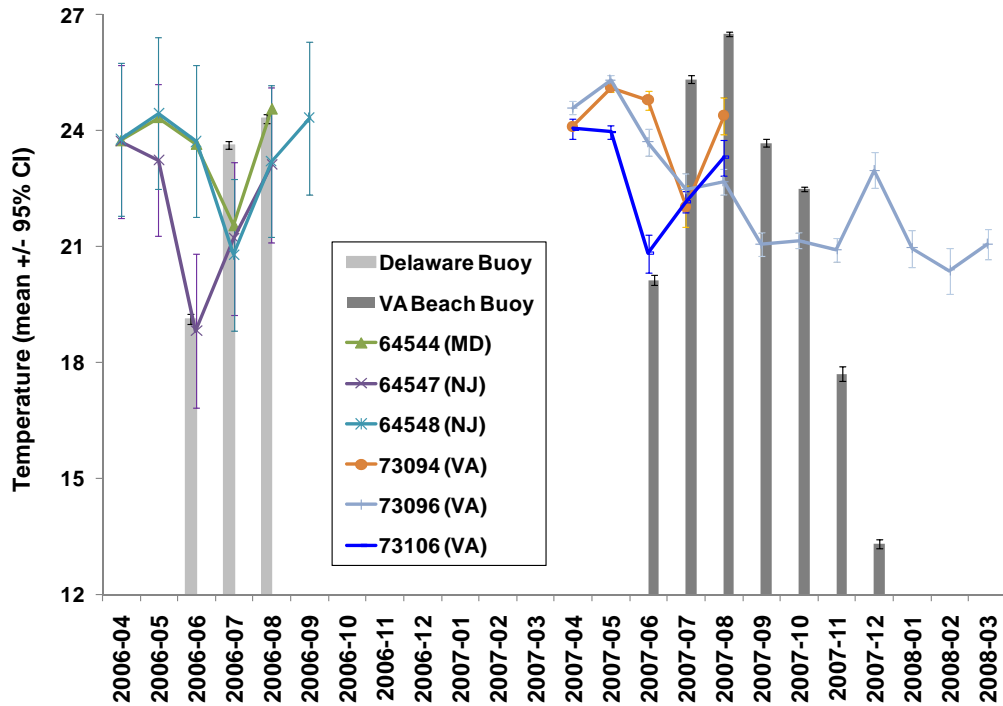




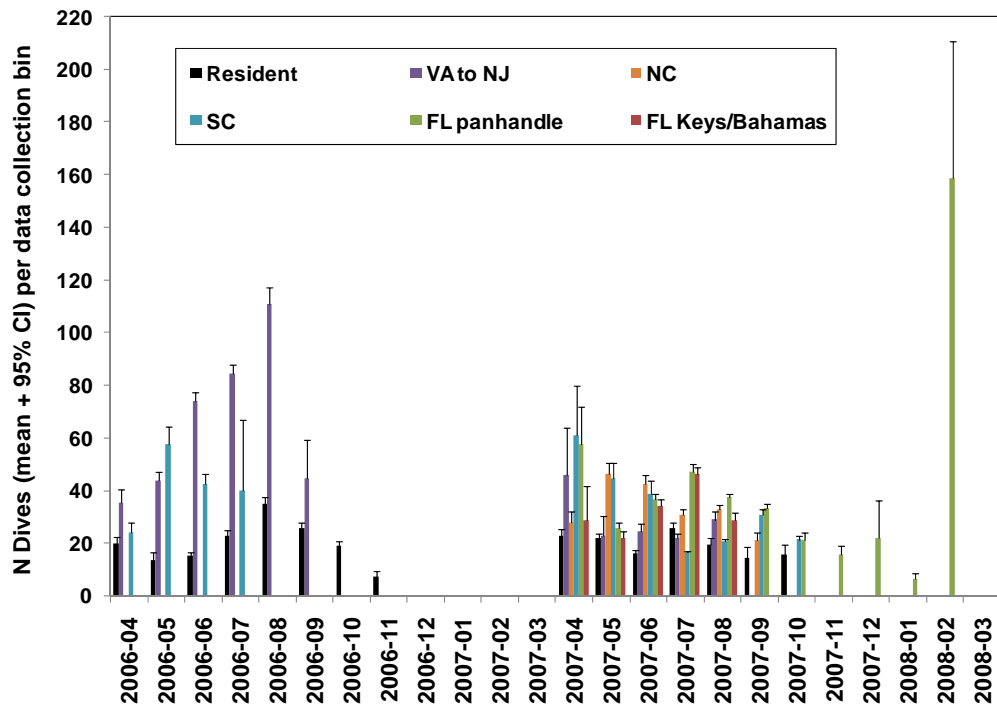
**Figure 5.5.** Monthly mean ( $\pm$  95% CI) transmitter temperature readings for resident male loggerheads and SST (mean  $\pm$  95% CI) temperature measured at the Cape Canaveral Buoy.



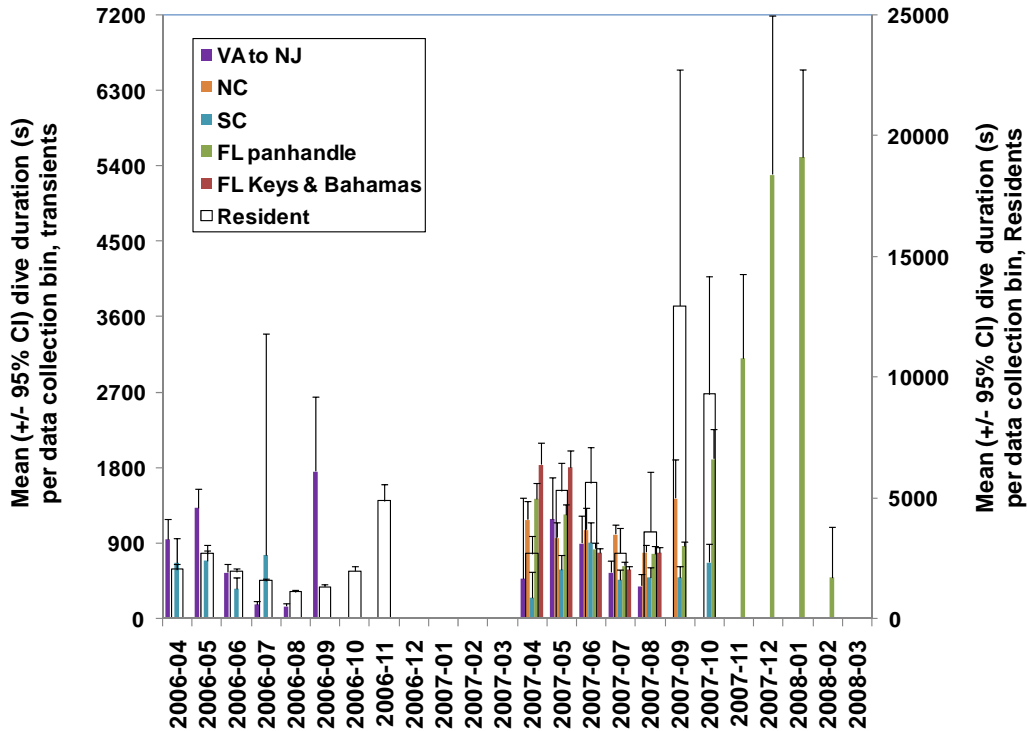
**Figure 5.6.** Comparison of monthly mean ( $\pm$  95% CI) transmitter temperature readings for transient male loggerheads while near Cape Canaveral, FL, (April and May) and while occupying five geographic dispersal zones upon emigrating away from Canaveral.



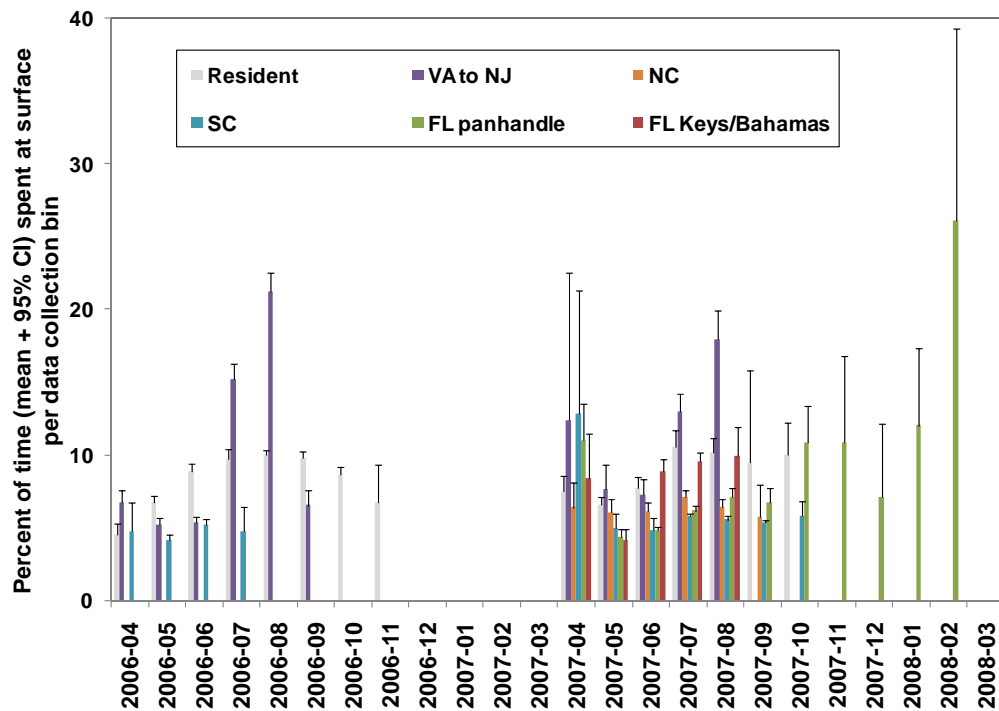
**Figure 5.7.** Disparity between transmitter (mean  $\pm$  95% CI) and sea surface (mean  $\pm$  95% CI) temperatures for six adult male loggerheads located between VA and NJ.



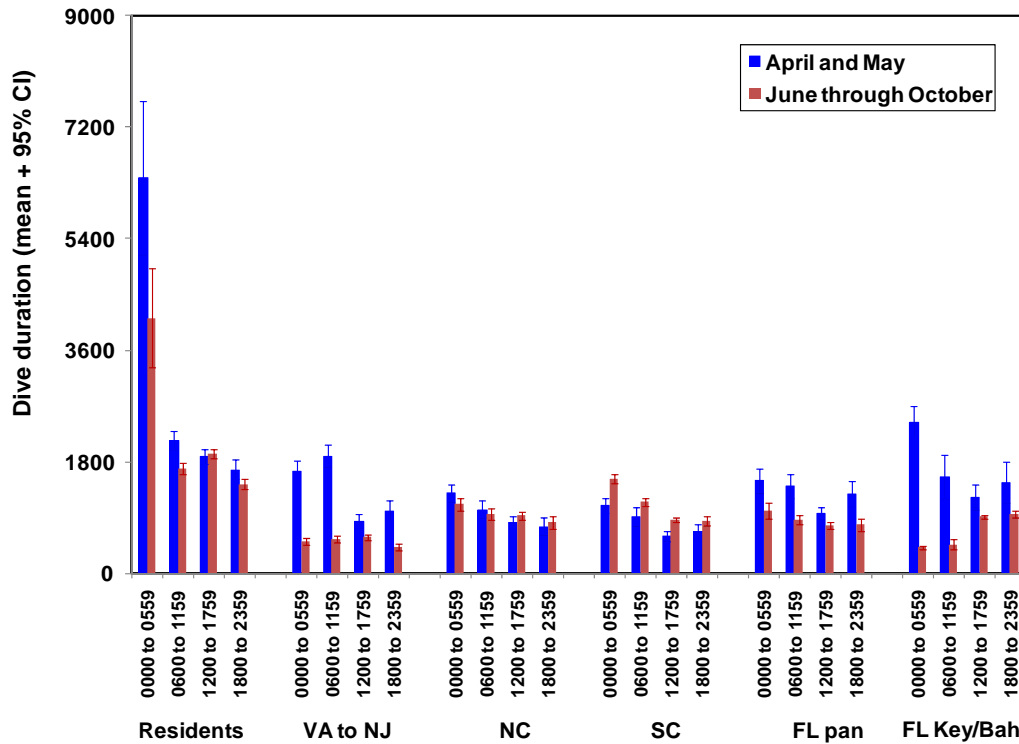
**Figure 5.8.** Temporal trends in the mean ( $\pm$  95% CI) number of dives made by adult male loggerheads during six hour data collection periods.



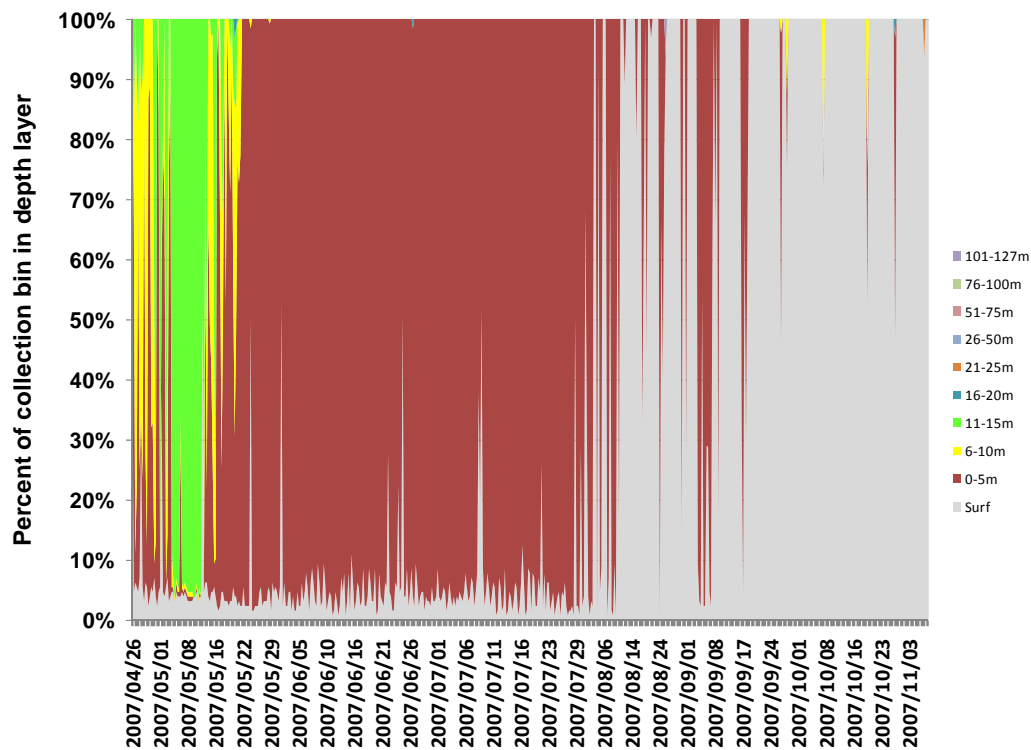
**Figure 5.9.** Temporal trends in mean (+ 95% CI) dive duration for adult male loggerheads during six hour data collection periods. Resident male data is provided on the second y-axis.



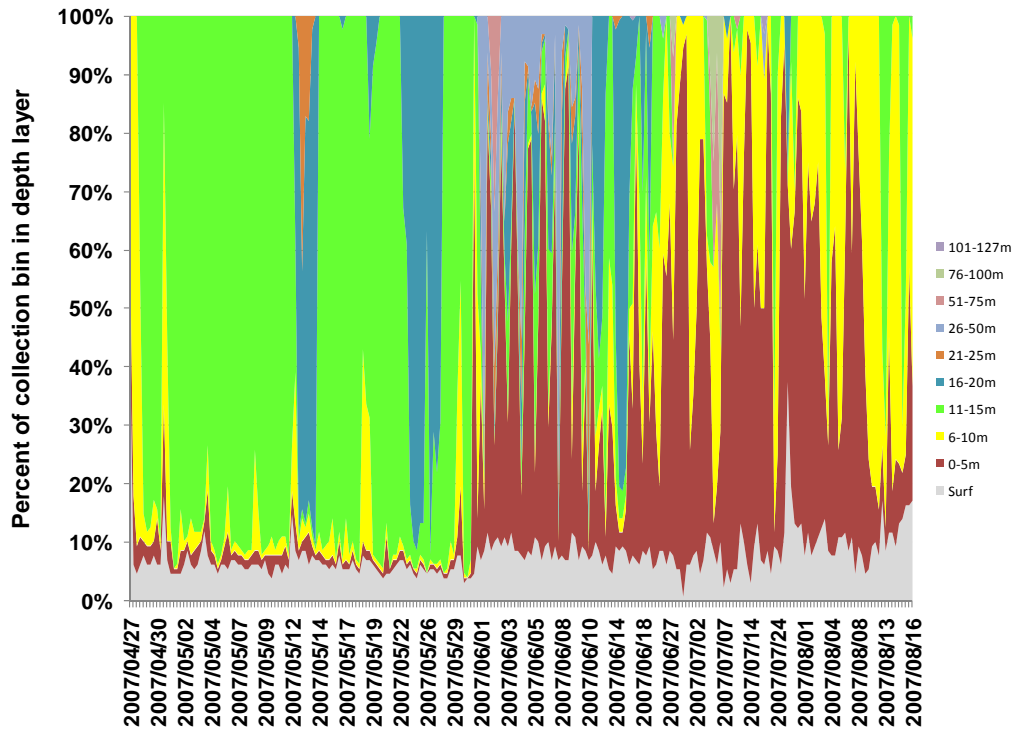
**Figure 5.10.** Temporal trends in the mean (+ 95% CI) percent of time spent at the sea surface by adult male loggerheads during six hour data collection periods.



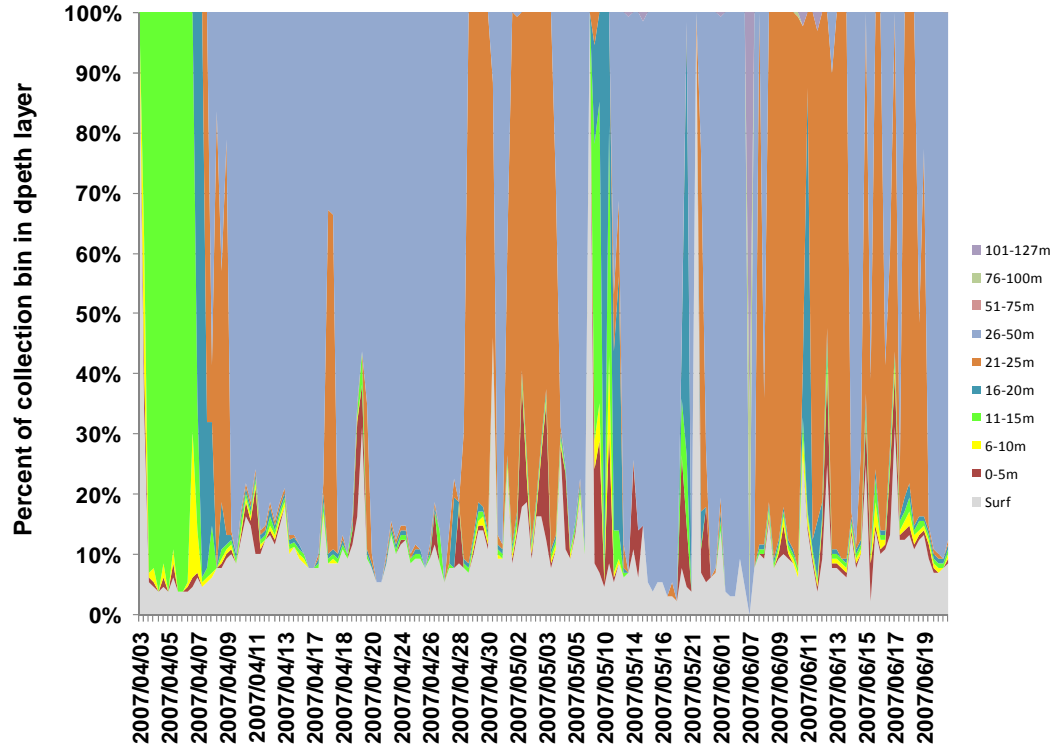
**Figure 5.11.** Diel trends in mean ( $\pm$  95% CI) dive duration for adult male loggerheads during six hour data collection periods.



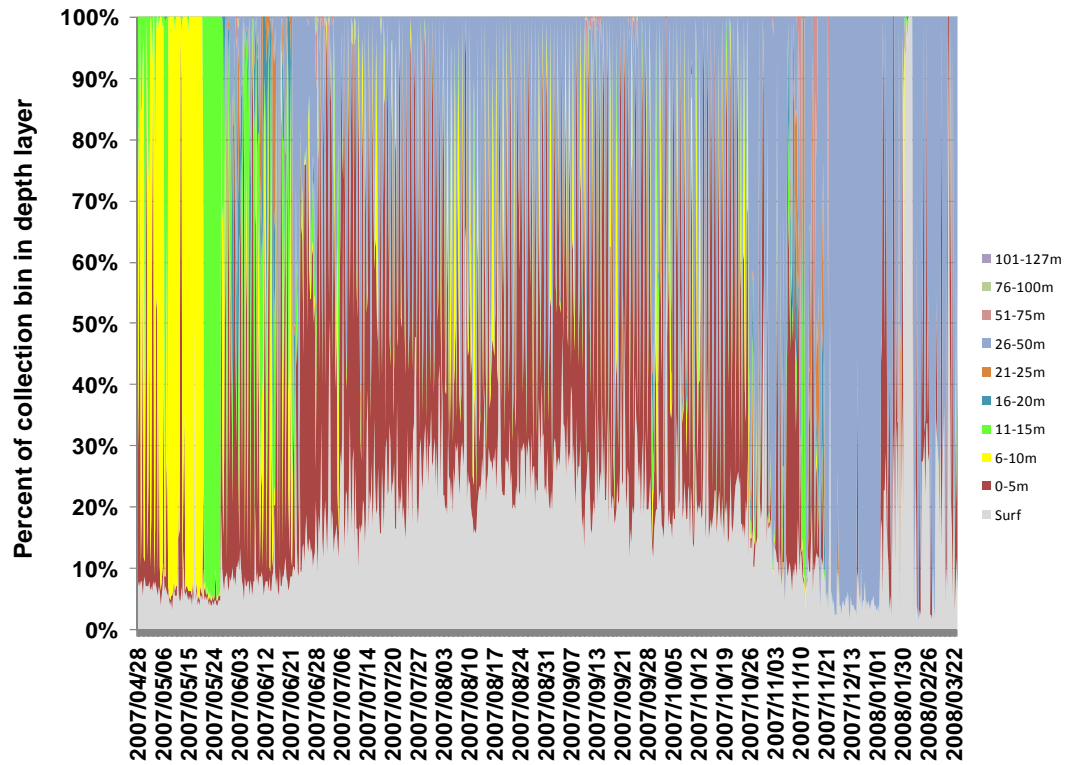
**Figure 5.12.** Dive depth distribution frequency for transient male ID#73093.



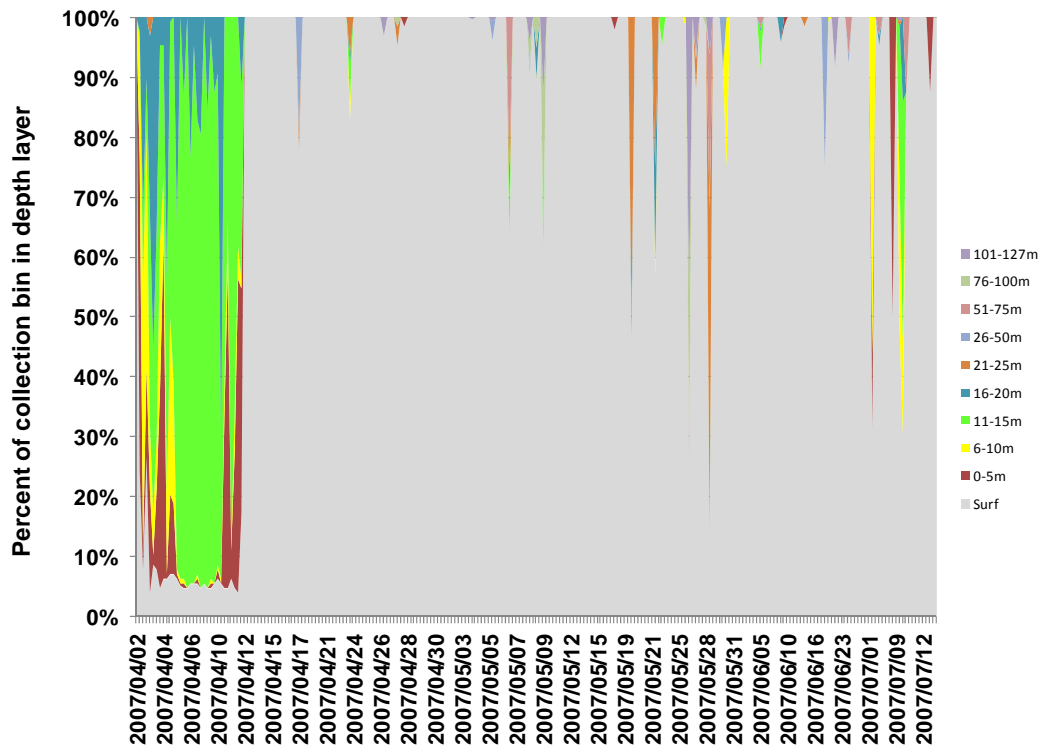
**Figure 5.13.** Dive depth distribution frequency for transient male ID#73094.



**Figure 5.14.** Dive depth distribution frequency for resident male ID#73095.



**Figure 5.15.** Dive depth distribution frequency for transient male ID#73096.



**Figure 5.16.** Dive depth distribution frequency for resident male ID#73097.

## Chapter 6 Blood mercury concentrations in juvenile and adult loggerheads (2001-2008).

### Introduction

The presence of heavy metals in the marine environment, their occurrence in marine organisms, and their role in the declining health of the world's oceans have attracted increased attention in recent decades. Anthropogenic input of several different heavy metals to aquatic ecosystems have caused concern; however, mercury (Hg) in particular is perhaps the most intensively studied and widely perceived contaminant threat to worldwide aquatic wildlife. Several studies have shown the capacity for Hg to accumulate in marine fish, seabirds, and mammals with such toxic effects as nervous, endocrine, and immune impairment and developmental, growth, and reproductive impacts (see Wolfe et al., 1998; Weiner et al., 2003; Scheuhammer et al., 2007 for reviews). In contrast, research into Hg effects on sea turtles remained largely deficient. Given their global conservation status as threatened or endangered species, identifying agents of impact to the recovery and long-term viability of sea turtle populations is paramount. Consequently, the occurrence and role of contaminants in sea turtle health and conservation have received greater attention in recent years. NOAA recovery plans for both the loggerhead (*Caretta caretta*) and Kemp's ridley (*Lepidochelys kempii*) sea turtle have mentioned both the lack of existing data among sea turtles (USFWS & NMFS, 1992) and the need to examine the impact of heavy metals in the recovery of populations (NMFS & USFWS, 2008).

Worldwide, Hg concentrations have been reported for individuals from multiple sea turtle populations. Researchers note the need for Hg data from tissues that can be easily and repeatedly sampled and have suggested using eggs (Sakai et al., 1995), blood (Day, 2003; Day et al., 2005) and the keratinaceous scutes covering the shell (Presti, 1999; Sakai et al., 2000a; Day et al., 2005) to monitor Hg in sea turtles. However, most Hg concentrations reported in the literature are from internal organs (liver, kidney) and tissue (skeletal muscle) thought to be most affected by high Hg burdens (Sakai et al., 2000a; Sakai et al., 2000b; Maffucci et al., 2005; Storelli et al., 2005). Furthermore, reports are typically biased towards samples obtained from debilitated, often dead, stranded turtles and small size classes, representing only the juvenile life stage. The result is a lack of concomitant data establishing baseline Hg concentrations in nonlethal sampling compartments and their relationship to internal tissues. Especially lacking are data for healthy individuals, particularly adult males which rarely return to shore and must be captured at sea.

Recent research in the southeast United States has focused on establishing optimal nonlethal sampling options for Hg monitoring in the loggerhead sea turtle and elucidating relationships between sampling compartments and internal tissues of interest. The loggerhead is the most abundant and common sea turtle species in coastal waters of the southeastern U.S. Following an oceanic juvenile stage, both adults and maturing (i.e. large juveniles) individuals inhabit coastal and estuarine waters where they opportunistically forage, predominantly on benthic prey. Foraging habitats are also susceptible to Hg pollution due to their proximity to coastal development and favorable sediment conditions for microbial-mediated conversion of inorganic Hg to the more bioavailable and toxic methylmercury (Weiner et al., 2003). Existing data, however, are scarce concerning Hg concentrations in loggerheads from U.S. waters. Presti

(1999) reported a mean blood Hg concentration of 14ppb in three live loggerheads from the Gulf of Mexico. More recently, mean blood Hg concentrations of 29ppb were reported for live loggerheads captured in near shore waters off the southeastern coast of the U.S. during 2001 (Day et al., 2005) and 2003 (Day et al., 2007). Currently, these studies serve as the only existing research documenting Hg concentrations in live loggerheads in the western Atlantic.

In the present study, we summarize five years of research describing blood Hg concentrations in loggerheads of the southeastern U.S. We revisit previously reported results (Day et al., 2005; Day et al., 2007) in light of additional data collected during the continuation of regional studies. We also present results from an additional sampling location for which Hg data has not been previously examined. Finally, blood Hg concentrations are examined in light of insight into turtle movement patterns provided by satellite tagging at two locations.

## Methods

### *Sample Collection and Field Processing*

Blood samples were obtained from free-ranging loggerhead sea turtles captured during research conducted by the South Carolina Department of Natural Resources (SCDNR). Samples included in this study were collected in 2001 and 2003-2008 by fishery-independent trawling aboard the R/V *Lady Lisa* and R/V *Georgia Bulldog* and the F/V *Miss Hilda* (2001) and F/V *Miss Tina* (2003). Samples collected during 2001 and 2003 cruises have been analyzed and presented previously (Day et al., 2005; Day et al., 2007) and are re-analyzed here as part of a larger data set. The data presented here for 2003 include only a subsample of the total 2003 data set described by Day et al. (2005) for which morphometric data were also available.

Samples obtained during 2001, 2003, and 2008 were collected as part of regional index-of-abundance studies sampling more than 500 trawl stations yearly from Winyah Bay, SC to St. Augustine, FL. Samples obtained during 2004-2007 were collected in conjunction with satellite tagging studies examining local and migratory movement of loggerheads along the Atlantic coast of the southeastern U.S. From 2004-2007, blood samples were collected from juvenile loggerheads captured in the Charleston, SC shipping channel and fitted with satellite tags. During 2006-2007 trawling efforts were also concentrated on the capture of loggerheads in the Port Canaveral, FL shipping channel. Samples obtained during Port Canaveral trawls constitute blood collected from satellite tagged adult male loggerheads, additional adults of both sexes and three juvenile turtles.

Briefly, turtles were captured using 19.8m trawl nets without turtle excluder devices. Trawl tow times were 10-30min depending on the year and sampling objective. Once onboard, turtles were measured, weighed, and fitted with a satellite tag (where applicable) before being released close to their capture location. Blood samples were taken from the dorsocervical sinus using a 21-gauge double-ended Vacutainer needle fitted to a 7ml heparinized Vacutainer blood collection tube (Becton-Dickinson, Franklin Lakes, NJ). Samples were immediately inverted five times and stored upright at -20°C both onboard and in the laboratory until processing. A separate sample was taken at each blood collection for immediate determination of packed cell volume (PCV), total protein and blood glucose concentration. Where necessary, the PCV was used to



convert total Hg (THg) concentrations measured in red blood cells to a whole blood THg concentration.

#### *Laboratory Techniques*

Total mercury concentrations were determined in collaboration with the National Institute of Standards and Technology (NIST), Charleston, SC. Archived blood was transferred from the original collection tubes to polypropylene sample vials using high-purity nitric acid (HNO<sub>3</sub>) rinsed glass pipets. All transfers were performed under a fume hood according to techniques designed to prevent sample contamination with ambient Hg. Only a small portion of the total blood volume collected for Hg analysis from each turtle was analyzed in this study. The remainder of each blood sample was archived in liquid nitrogen freezers according to NIST specimen archive protocols and is available for future heavy metal contaminant studies.

Loggerhead whole blood and red blood cells (where applicable) were analyzed via cold vapor atomic fluorescence spectroscopy (CV-AFS) on a PSA 10.025 Millennium Merlin (PS Analytical, Orpington, UK). In general, 0.25g of tissue were digested in 1ml high-purity HNO<sub>3</sub> and 0.25ml hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) in quartz microwave digestion vessels using a CEM Discover Benchmate microwave (CEM, Matthews, NC) set to maximum values of 150°C and 230psi. Typical digestion conditions reached approximately 100°C and 230psi. Following digestion, vessels were vented under a fume hood for five minutes and digests were quantitatively diluted by a factor of 100 to 150 in HNO<sub>3</sub> rinsed polypropylene sample tubes. Equal volumes of this original dilution were quantitatively delivered to separate acid rinsed sample tubes, designated “unspiked” and “spiked” splits respectively. Spiked splits received a quantitative standard addition of NIST SRM 3133 Mercury Spectrometric Solution while unspiked splits received an equivalent volume of a 2.5% HNO<sub>3</sub> - 1% hydrochloric acid (HCl) “dummy” spike. Unspiked and spiked splits were analyzed in succession and in triplicate after reduction to Hg(0) with stannous chloride (SnCl<sub>2</sub>).

Two hundred twenty-nine blood samples were analyzed in 28 batches from July to December 2008. A sample batch generally consisted of 10 blood samples, a NIST SRM 966 Trace Metals in Bovine Blood digest, and a procedural blank. Raw data from spiked and unspiked splits were transferred to Microsoft Excel and used to generate a two point standard addition curve for each sample. The concentration of unknown Hg in the sample was calculated from the equation for the standard addition curve. Analytical methods for the determination of THg in the 2001 and 2003 samples differed from those used here and have been previously described (Day et al., 2005; Day et al., 2007).

#### *Statistical Analysis*

Statistical analyses were performed on R version 2.5.0 (R Core Team, Vienna, Austria) and Minitab 15® (MiniTab Inc., State College, PA) statistical software. Where necessary, Shapiro-Wilk and Bartlett’s tests were used to ensure data met assumptions of normality and homoscedasticity of variance respectively. Nonparametric Kruskal-Wallis (hereafter abbreviated K-W) and Wilcoxon tests were used where data deviated from normality or equal variance.

All concentrations reported here are expressed as wet weight THg, reported in parts-per-billion (ppb, ng/g)  $\pm$  1 SD. Turtle length measurements are notch-notch straight carapace length (SCLmin, cm) except where noted.

## Results

Data for 313 loggerheads were available for statistical analyses. Thirty samples collected during 2001 and 54 samples collected during 2003 were included in these analyses. In addition, 229 loggerhead blood samples analyzed during 2008 included: 33 Charleston channel satellite-tagged samples, 46 additional Charleston channel samples, 29 Port Canaveral satellite-tagged samples, 39 additional Port Canaveral samples, and 82 samples from the 2008 regional index of abundance study. Total mercury concentration in loggerhead whole blood ranged from 1.48ppb to 408.40ppb and averaged  $34.39 \pm 35$ ppb for all individuals sampled from 2001-2008.

Mercury correlations with body size have been reported for some tissues in sea turtles species and the data presented here incorporate samples obtained from turtles of various sizes. Turtle size, measured as SCLmin, differed between sites (K-W,  $H=60.006$ ,  $p=2.89E-12$ ); however, there was no significant overall relationship between turtle body size and blood THg (Kendall's tau=0.0013,  $p=0.971$ , Figure 6.1A). Port Canaveral turtles had the largest mean size,  $85.1 \pm 13.1$ cm, while turtles from northern Florida waters had the smallest mean size,  $67.6 \pm 10.8$ cm. Turtles were grouped by SCLmin into the following size classes: <65cm, 65.1-75cm, 75.1-85cm, 85.1-95cm, and >95cm. These classes also correspond to recognized delineations in loggerhead reproductive status in the North Atlantic. In this study, loggerheads <75cm were considered juveniles, those between 75.1cm and 85cm were considered "maturing" individuals (i.e. approaching transition to reproductive maturity), and loggerheads >85.1cm were considered adults (i.e. reproductively active). Taken together, blood THg concentrations were not significantly different between any size class (K-W,  $H=4.121$ ,  $p=0.389$ ). Mean blood THg was highest in the 85.1-95cm size class ( $45.39 \pm 59.2$ ppb). However, blood THg was lowest ( $27.23 \pm 18.5$ ppb) in the next largest size class, turtles >95cm. Consequently, blood THg was also not significantly different between the three reproductive stages (K-W,  $H=2.325$ ,  $p=0.507$ ). Additionally, concentrations were not statistically different between sexes (K-W,  $H=0.240$ ,  $p=0.623$ , Figure 6.2A) for all animals sampled. Overall, males had a mean blood THg concentration of  $37.16 \pm 47.8$ ppb while the mean for females was  $33.22 \pm 26.6$ ppb.

Samples included in this study were also obtained from a large geographic area. Thus, the potential exists for latitudinal variability in blood THg concentrations based on factors influencing local Hg availability in the environment. Turtles were grouped into the following classes based on capture location to test for a geographic effect on blood THg concentration: Winyah Bay, SC to Charleston, SC, Charleston to Savannah, GA, Savannah to Brunswick, GA, Brunswick to St. Augustine, FL, and Port Canaveral, FL. Overall, blood THg concentrations were statistically different between geographic classes (K-W,  $H=17.870$ ,  $p=0.0013$ , Figure 6.3). Subsequent nonparametric Dunn-Bonferroni pairwise comparisons showed statistically significant differences in blood THg concentrations for the following comparisons: Port Canaveral vs. Brunswick-St. Augustine ( $p < 0.0001$ ), Port Canaveral vs. Winyah Bay-Charleston ( $p=0.011$ ), and Winyah Bay-Charleston vs. Brunswick-St. Augustine ( $p=0.015$ ). Port Canaveral

loggerheads had the highest mean blood THg,  $52.71 \pm 63.1$ ppb, while turtles captured from Brunswick, GA to St. Augustine, FL had the lowest mean blood THg,  $26.21 \pm 29.0$ ppb.

#### *Regional Index of Abundance Study*

The total sample size for the regional index of abundance study consisted of 166 loggerhead blood samples, including: 30 samples collected during 2001, 54 samples collected during 2003, and 82 samples collected during 2008. Regional study turtles ranged in size from 50.2cm to 94.9cm and included animals from all reproductive stages (Fig. 6.4). There were no overall statistical differences in blood THg concentration due to size (K-W,  $H=118.595$ ,  $p=0.792$ , Figure 6.1B) or reproductive status (K-W,  $H=0.401$ ,  $p=0.818$ ) across all years. Irrespective of size, there were also no significant yearly (K-W,  $H=1.065$ ,  $p=0.587$ , Figure 6.5) or sexual (K-W,  $H=0.919$ ,  $p=0.337$ , Figure 6.2B) effects on blood THg concentrations across the region. Over the three years sampled, mean blood THg concentrations decreased yearly from  $31.01 \pm 32.1$ ppb during 2001 to  $27.96 \pm 16.1$ ppb during 2008. Over all years, male loggerheads had a higher mean blood THg concentration,  $32.48 \pm 26.1$ ppb, than females,  $26.86 \pm 15.8$ ppb.

Geographically, only minor overall latitudinal variability was apparent across years within the regional study (Figure 6.6). Minor variability was also evident between subregions within a given year (Figure 6.6). Mean blood THg concentrations for all subregions ranged from  $21.13 \pm 17.0$ ppb to  $36.34 \pm 49.2$ ppb over the three years sampled. In general, mean blood THg concentrations decreased latitudinally from South Carolina to Florida waters. Turtles captured between Brunswick, GA and St. Augustine, FL had the lowest mean THg concentration in both 2003 and 2008. A similar result was evident in 2001 when the capture of a single individual with a high blood THg concentration (188.26ppb) was removed. Overall, however, there was a significant difference in THg concentration between subregions (K-W,  $H=9.570$ ,  $p=0.022$ ). Dunn-Bonferroni pairwise comparisons revealed significant differences between blood THg concentrations for the following locations: Charleston-Savannah vs. Brunswick-St. Augustine and Savannah-Brunswick vs. Brunswick-St. Augustine.

#### *Satellite-Tagging Study*

Sixty-two loggerheads were fitted with satellite tags during concentrated sampling in the Charleston, SC ( $n=33$ ) and Port Canaveral, FL ( $n=29$ ) ship channels from 2004-2007. Sampling targeted juvenile animals of both sexes in the Charleston area and adult male loggerheads in the Port Canaveral channel. Tracking also indicated the presence of both “transient” and “resident” turtles in both locations. There was no indication of differences in blood THg concentration between animals tagged in either location (K-W,  $H=0.044$ ,  $p=0.832$ , Figure 6.7), however, the standard deviation of THg concentrations among Port Canaveral-tagged turtles was rather large. Blood THg averaged  $29.99 \pm 13.2$ ppb for turtles tagged in the Charleston ship channel and  $46.9 \pm 76.0$ ppb for those tagged in Port Canaveral. Additionally, there was no statistical difference in blood THg between transient and resident individuals in either Charleston (ANOVA,  $F=0.003$ ,  $p=0.956$ , Figure 6.8) or Port Canaveral (K-W,  $H=0.137$ ,  $p=0.710$ , Figure 6.8). Among Charleston-tagged turtles, residents ( $30.0 \pm 13.8$ ppb) had a slightly greater blood THg concentration than transients ( $29.7 \pm 10.4$ ppb); however, only six Charleston channel turtles were classified as transient. Blood THg among Port Canaveral-tagged individuals was greater in transient turtles ( $50.41 \pm 97.1$ ppb) than in residents ( $43.99 \pm 42.3$ ppb). There was also no significant sex effect on blood THg among Charleston-tagged turtles (ANOVA,  $F=3.090$ ,

$p=0.088$ , Figure 6.2C). Blood THg averaged  $22.57 \pm 10.7$ ppb among male Charleston-tagged loggerheads and  $32.29 \pm 13.4$ ppb among females.

It should also be of note whether blood THg concentrations for satellite-tagged loggerheads are similar to those for other individuals caught at each tagging locale and are likely indicative of the extent of exposure to the overall population at each site. Charleston-tagged turtles were, therefore, compared with other juvenile and “maturing” size ( $n=31$ ) and adult turtles ( $n=15$ ) captured in the Charleston channel during the same sampling events. Blood THg among Charleston-tagged turtles was not statistically different from these other Charleston-captured turtles (K-W,  $H=0.873$ ,  $p=0.350$ ). Conversely, blood THg concentration among Port Canaveral-tagged turtles was statistically different from other loggerheads captured in the Port Canaveral channel (K-W,  $H=4.365$ ,  $p=0.036$ ). In Port Canaveral, the mean blood THg concentration was greater among non-satellite tagged turtles  $57.01 \pm 52.2$ ppb than satellite tagged individuals ( $46.9 \pm 76.0$ ppb).

#### *Charleston Channel Study*

Although sampling events in the Charleston, SC shipping channel were concentrated on the capture of juvenile turtles, additional turtles of all size classes and both sexes were also sampled. The blood THg concentration of turtles captured during targeted Charleston channel sampling was consistent with that of turtles captured in South Carolina waters during the regional index of abundance study (Wilcoxon test,  $W=2304$ ,  $p=0.291$ ) despite a significant difference in the mean size of each group (Wilcoxon test,  $W=1849$ ,  $p=0.003$ ). Additionally, there was no significant difference in blood THg concentration between satellite-tagged and other Charleston turtles (see above).

Seventy-nine total samples were analyzed for turtles captured in the Charleston channel. Overall, there was no significant relationship between turtle body size and blood THg concentration among Charleston channel turtles (Kendall’s regression,  $p=0.230$ , Figure 6.1C). There were also no significant differences between the size (K-W,  $H=64.37$ ,  $p=0.568$ ) or reproductive classes (K-W,  $H=0.312$ ,  $p=0.855$ ) outlined above. There was, however, a significant sex effect on blood THg concentration among all Charleston channel samples (K-W,  $H=4.969$ ,  $p=0.025$ , Figure 6.2D). Females from the Charleston channel had a greater mean blood THg ( $30.77 \pm 14.3$ ppb) than males ( $21.5 \pm 13.3$ ppb).

The duration of the sampling season in the Charleston channel also allowed for a seasonal comparison of THg in loggerhead blood. Ten turtles were randomly selected from all individuals captured during May and August in both 2004 and 2005 to test for seasonal differences. During 2004, mean blood THg was significantly different between these two months (ANOVA,  $F=7.643$ ,  $p=0.012$ ), with the mean THg for May ( $39.18 \pm 15.9$ ppb) being greater than August ( $22.05 \pm 11.5$ ppb). Conversely, there was no significant difference between May and August means during 2005 (ANOVA,  $F=0.006$ ,  $p=0.936$ ). Blood THg concentrations within the month of May were also significantly different between the two years (ANOVA,  $F=7.893$ ,  $p=0.011$ ) while those within the month of August were not (ANOVA,  $F=0.048$ ,  $p=0.827$ ).

### *Port Canaveral Study*

The mean blood Hg concentration of non-satellite tagged turtles was greater than that for the tagged male turtles within Port Canaveral (see above). Non-tagged Port Canaveral turtles consisted of both male ( $n=14$ ) and female ( $n=25$ ) turtles. However, there was no significant effect on blood THg due to sex (K-W,  $H=0.9334$ ,  $p=0.334$ , Figure 6.2E) in these turtles. Additionally, non-tagged turtles also consisted of juvenile ( $n=18$ ), “maturing” ( $n=4$ ), and adult ( $n=17$ ) size turtles while only adult turtles were fitted with satellite tags. However, there was also no significant relationship between blood THg and size among non-tagged turtles (Kendall’s regression,  $p=0.743$ ). Thus, blood THg was not significantly different between size (K-W,  $H=62.539$ ,  $p=0.385$ , Figure 6.1D) or reproductive classes (K-W,  $H=8.0E-4$ ,  $p=0.977$ ; juvenile and maturing size animals combined due to sample size) in these turtles. Port Canaveral sampling also took place over two field seasons, 2006 and 2007 so there was potential for variability between years in animals captured during sampling in this location. Blood THg concentrations, however, were not significantly different between years (Wilcoxon test,  $W=551$ ,  $p=0.247$ ).

### **Discussion**

In general, blood THg concentrations reported here are consistent with results from three previous studies documenting Hg in the blood of live loggerhead turtles in the western Atlantic. Reported values in these studies range from 14 to 29ppb (Presti, 1999; Day et al., 2005; Day et al., 2007). Mean THg concentration of all loggerheads in the current study was  $34.3 \pm 34.9$ ppb while the mean THg concentration of only the 229 loggerheads not previously reported (i.e. those not included in the Day et al. 2005 and 2007 studies) was  $36.13 \pm 38.6$ ppb. Mercury concentrations in sea turtle blood are rare in the literature. However, additional blood Hg values have also been reported for Kemp’s ridley sea turtles along the U.S. Atlantic coast and in the Gulf of Mexico. Orvik (1997) and Presti (1999) reported mean blood Hg concentrations of 18 and 27ppb respectively for Kemp’s ridleys from Texas waters. Maximum reported values in those studies were 67 and 87ppb respectively. Wang (2005) reported mean Hg concentrations of 16ppb for Kemp’s ridleys from U.S. waters of the Gulf of Mexico and 69ppb for individuals captured near the historic Kemp’s ridley nesting beach at Rancho Nuevo, Mexico. Maximum reported concentrations for those populations were 179 and 145ppb respectively. Finally, Innis et al. (2008) reported a mean blood Hg concentration of 24ppb and a maximum concentration of 43ppb in juvenile cold-stunned Kemp’s ridleys from Cape Cod, MA.

The greatest blood THg concentration previously reported for an Atlantic loggerhead was 188ppb for an individual captured from Georgia waters during 2001 regional sampling (Day et al., 2005). In the current study, three turtles had blood Hg concentrations  $>188$  ppb (range= $204.12 - 408.40$  ppb). All three turtles were captured during sampling in the Port Canaveral shipping channel and all three were greater than 81.1cm SCLmin in size. Interestingly, however, some of the lowest blood Hg concentrations were also found among turtles captured at Port Canaveral. Four Port Canaveral turtles had blood Hg concentrations of four ppb or lower, including the lowest THg concentration measured in this study (1.48ppb), and all four turtles were greater than 86.9cm SCLmin. The greatest blood Hg for any turtle captured outside of Port Canaveral was 108.15ppb for a 66.1cm juvenile captured off Edisto Island, SC and the lowest concentration outside Port Canaveral was 1.73ppb for a 98.8cm SCLmin adult male loggerhead captured in the Charleston shipping channel.

These concentration/size discrepancies in individual turtles highlight the overall lack of correlation between turtle size and Hg concentration in both the individual studies presented here and the complete data set (2001-2008). Mercury has previously been shown to accumulate in larger (i.e. potentially older) individuals within a species, mostly among fish (Weiner and Spry, 1996; Weiner et al., 2003). This finding has often been confirmed through examination of muscle tissue or whole body Hg burdens, therefore comparable data for the blood compartment are rare. Fish also lack accompanying routes for Hg sequestration and elimination seen among other vertebrates (Weiner et al., 2003) such as feathers, hair, or scutes. Thus, any blood vs. size correlation among sea turtles should be more difficult to establish than for fish. Sea turtle studies must rely on size data to establish such correlations given the difficulty in estimating age in these organisms. Few previous studies have examined relationships between body size and blood THg in sea turtles, especially over large size ranges. Wang (2005) reported an insignificant ( $R^2=0.0001$ ) correlation between blood Hg concentration and SCL in Kemp's ridley turtles from Texas waters, however the sample size consisted of only juvenile turtles (<60cm SCL). Innis et al. (2008) reported similar results for juvenile Kemp's ridleys from Massachusetts ( $R^2=0.44$ ,  $p>0.12$ ) and suggested a lack of larger turtles may have contributed to these results. Conversely, Day et al. (2005) reported a significant relationship ( $R^2=0.17$ ,  $p=0.016$ ) between body mass and blood THg concentration for loggerhead turtles collected during the 2001 sampling preceding this study. Both juvenile and adult size turtles were included in that study.

Significant Hg/size correlations have also been reported for Hg in sea turtle scutes. For example, Presti et al. (1999) reported a significant scute Hg vs. size relationship among black turtles (*Chelonia mydas agassizii*), although the sample size was small and the relationship stronger in larger (>42cm SCL) compared to smaller turtles. Day et al. (2005) also reported a significant scute THg concentration vs. body mass ( $R^2=0.188$ ,  $p=0.012$ ) relationship for loggerheads. The current study offered the opportunity to establish a better blood Hg/body size correlation given the larger sample size and the addition of a large number of individuals in bigger size classes. However, nonparametric regression revealed no overall significant relationship between either blood THg and SCLmin or blood THg and body mass (Kendall's tau=0.006,  $p=0.864$ ). Additionally, the blood THg vs. SCLmin relationship was not significant for any single size class (Kendall's regression  $p>0.19$  for all size classes). Blood is generally considered to be a short-term depository for Hg ingested via the diet and is, therefore, influenced by changes in foraging behavior. Furthermore, dietary Hg is ultimately in a complex equilibrium with Hg pools in other body compartments. Thus, the blood THg concentration at any single sampling point is confounded by multiple variables. It remains possible that the inclusion of more individuals in all size classes could better define the relationship. However, considering the large geographic range included in this study, additional information regarding rangewide differences in turtle ecology and life history status, foraging behavior, movement patterns, Hg concentrations in local prey species, and Hg availability to turtle populations must also be obtained.

Fifty five percent of all adult turtles (i.e.  $\geq 85.1$ cm SCLmin) in this study were captured during sampling in the Port Canaveral shipping channel. Thus, confirmation of geographic differences in blood THg is interesting, particularly given the lack of correlation between blood THg and turtle body size or body mass. Overall, blood THg varied significantly between sites and nonparametric Dunn-Bonferroni post-hoc comparison revealed significant differences in blood THg for three site comparisons. Dunn-Bonferroni comparisons, however, assign ranks to raw

data and tested for significant differences between the median ranks across location. A comparison of actual blood THg means between locations suggests THg concentrations are highly similar across the sampling region outside Port Canaveral. Blood THg means for the four geographic classes outside Port Canaveral ranged from 26.21-29.47ppb. Although the Dunn-Bonferroni results suggested differences in blood THg between Winyah Bay-Charleston and Brunswick-St. Augustine turtles, the mean THg concentration of animals sampled in these two regions differed by only 3.26ppb. The remaining significant Dunn-Bonferroni comparisons both included the Port Canaveral sampling area (Port Canaveral vs. Brunswick-St. Augustine and Port Canaveral vs. Winyah Bay-Charleston) and the mean THg concentration for Port Canaveral turtles was 23ppb greater than the next highest geographic class (Winyah Bay-Charleston). Additionally, the lowest recorded mean THg concentration was for turtles captured within the next closest region to Port Canaveral (Brunswick-St. Augustine). These results suggest the Port Canaveral area may receive higher Hg inputs compared with other sampling areas in the southeast region. Alternatively, a difference in the ecology of turtles in the Port Canaveral area may result in greater Hg exposure compared to turtles in other sampling locations. We briefly examine both possibilities below.

The relatively stable blood Hg concentrations between years and within individual subregions seen in the regional study suggest Hg availability to the marine food web in the southeast remained relatively stable from 2003-2008. Alternatively, it is possible any change in Hg input during this time is not yet manifested in loggerheads in this region. Nonetheless, mean blood THg concentrations for both juveniles and maturing/adult turtles from Port Canaveral were almost 2X greater than mean concentrations for juveniles or maturing/adult turtles captured in either South Carolina or Georgia waters. Maps generated by the Mercury Deposition Network (MDN, <http://nadp.sws.uiuc.edu/mdn/maps>) suggest both the concentration and amount of wet Hg deposition from South Carolina to Florida have remained relatively stable since at least 2000. These same maps, however, also show higher annual Hg input to peninsular Florida compared with input to South Carolina or Georgia over the same period. Furthermore, according to MDN data, portions of peninsular and south Florida consistently receive some of the greatest amounts of annual wet Hg deposition nationwide. Such high deposition occurs despite data showing past anthropogenic Hg emissions within the state of Florida are similar to the national average (<http://epa.gov/ttn/atw/combust/utiltox/stxstate2.pdf>).

Globally, up to 2/3 of the current atmospheric Hg pool has been attributed to anthropogenic sources (Mason et al., 1994; Lindberg et al., 2007). Furthermore, depending on the species composition, anthropogenic Hg emissions have been shown to be deposited in significant amounts in localized regions close to emission sources as well as transported over large distances in a truly global pool (Schroeder and Munthe, 1998; Weiner et al., 2003; Valente et al., 2007). Thus, the Hg pool at any locale can be composed of differing amounts of Hg from varying anthropogenic sources: local, regional, and global. In the state of Florida, Hg from localized anthropogenic sources has been shown to account for a sizeable portion of wet deposition Hg input to both the Florida Everglades and south Florida (Dvonch et al., 1999; Guentzel et al., 2001; Dvonch et al., 2005). The south Florida region identified in these studies was generally from Lake Okechobee southward and Guentzel et al. (1995) reported Hg deposition to north and central Florida was 30-50% less than in south Florida. However, it is not unreasonable to

suppose that significant local Hg input, combined with other factors, could contribute to increased contamination near Port Canaveral compared with more northern sites sampled.

Anthropogenic Hg emissions come from numerous sources, including: the combustion of coal and other fossil fuels; municipal, hazardous, and medical waste incineration; and paper mills, mining activities, and production of certain industrial products (Weiner et al., 2003). However, coal combustion is the largest contributor to both global ( $\approx 60\%$  of emissions) and U.S. ( $\approx 30\%$ ) anthropogenic emissions (Swain et al., 2007). Coal combustion currently produces at least 30 to 40% of the energy used in South Carolina, Georgia and Florida according to the Department of Energy ([www.eia.doe.gov](http://www.eia.doe.gov)). These states also consume energy at a rate higher than the national average ([www.eia.doe.gov](http://www.eia.doe.gov)). While South Carolina and Georgia both possess several coal-fired power plants located in coastal counties, one of the largest (by megawatt output) coal-fired plants in Florida (<http://www.dep.state.fl.us/Air/permitting/construction/powergen.pdf>) is located approximately 70km west of Port Canaveral. Additionally, Port Canaveral lies approximately 90km from the heavily populated and industrialized Orlando, FL area. Thus far, published studies of Hg cycling and deposition in Florida do not include any data collected near the Port Canaveral area. However, previous studies have shown that anthropogenic emissions can, indeed, be deposited over more local scales, especially when comprised of larger amounts of particulate mercury or Hg(II) species compared to predominantly elemental Hg<sup>0</sup> emissions (Schroeder and Munthe, 1998; Seigneur et al., 2003; Weiner et al., 2003). These more reactive Hg species have atmospheric residence times of hours to weeks and are thought to be deposited over much smaller (i.e. hundreds of km) geographic scales than Hg<sup>0</sup> (Seigneur et al., 2003). In the U.S., sizeable contributions of local and regional scale Hg emissions to overall deposition have been reported for some sites in the Great Lakes (Lynam and Keeler, 2005), New York (Bookman et al., 2008) and Florida (Facemire et al., 1995; Dvonch et al., 1999; Guentzel et al., 2001; Dvonch et al., 2005). Valente et al. (2007) summarized data showing deposition of Hg from local sources typically constitutes a greater percentage of overall deposition in urban areas. It is conceivable, therefore, that anthropogenic Hg emissions from local sources have contributed to greater Hg input to the Port Canaveral area. The extent to which such increased deposition in the Port Canaveral area would ultimately be manifested in loggerhead blood and tissues, however, requires further research.

The increased rate and concentration of Hg input to Florida, compared to South Carolina and Georgia, is also most certainly a result of weather-related effects on Hg deposition. Mercury deposition is known to be influenced by atmospheric weather patterns and past research shows significant contributions to Florida's Hg deposition due to both local and global sources. Preliminary analysis of just a single meteorological factor, wind direction, at seven sites within our study region revealed subtle differences that may help to explain at least a portion of the variation in loggerhead blood Hg concentration between locations. Data recorded by near shore buoys located at North Inlet/Winyah Bay, SC, Capers Island, SC, Ft. Pulaski/Savannah, GA, Sapelo Island, GA, Fernandina Beach, FL, and Port Canaveral over a 2-3 year period (2006-2008, National Data Buoy Center, [www.ndbc.noaa.gov](http://www.ndbc.noaa.gov)) reveal distinct differences in wind direction patterns across the southeast region. Mean annual wind direction recorded at southern buoys was generally southeasterly or south southeasterly. Conversely, buoys located from Sapelo Island northward recorded more southwesterly seasonal and annual mean wind directions.



The wind directions recorded by these buoys are sea level wind directions. Conceivably, though, differences in atmospheric winds between sites may influence Hg deposition on a local scale in addition to influencing more long-range Hg transport. For example, studies have linked an increase in Hg deposition to south Florida during summer months to increased summer rainfall (Guentzel et al., 2001; Fulkerson and Nnadi, 2006). Guentzel et al. (2001) hypothesized that strong southeasterly summer winds served to supply steady amounts of atmospheric reactive gaseous Hg, which subsequently falls during heavy summer thunderstorms initiated by these same winds. It is also possible that high-elevation atmospheric winds over the open ocean or continental U.S. could entrain additional anthropogenic emissions from more distant sources, ultimately resulting in their deposition within Florida. Indeed, Guentzel et al. (2001) reported greater than 50% of wet deposition Hg during summertime rain events in south Florida came from long-range sources. Over a more localized scale, a weather pattern dominated by southeasterly winds may also serve to decrease dilution of Hg in Port Canaveral waters by limiting circulation with the open ocean. The Port Canaveral lagoon is known to experience low tidal ranges and a lack of tidal flushing. Perhaps, southeasterly winds, when coupled with low tidal exchange, could lead to increased methylation efficiency within Port Canaveral waters and increased exposure to loggerheads and other fauna in surrounding areas. Srinivasan and Swain (2007) suggested tides and local weather patterns influence retention of high levels of copper associated with boat antifouling paints within the Port Canaveral lagoon. Thus, further examination of the influence of such factors as wind direction, tidal amplitude, and frequency, seasonality, and amount of rainfall (given the importance of wet Hg deposition to total deposition) may provide further insight regarding differences in Hg input over the geographical range sampled in this study.

An alternative hypothesis for the greater blood Hg concentrations seen at Port Canaveral involves the ecology of sea turtles found in near shore waters of the Port Canaveral area. The Port Canaveral channel and surrounding waters have been widely studied since the early 1970's due to the high concentration of sea turtles found there. In addition to large adult aggregations, the existence of large numbers of juvenile and maturing size individuals has been recorded in Port Canaveral waters over much of the year. Henwood (1987b) reported juvenile size turtles (carapace length <69cm) comprised 83% of the loggerheads captured during five trawling studies in Port Canaveral waters. Similarly, Schmid (1995) reported juvenile loggerheads comprised 80% of turtles captured during tagging studies from 1986-1991. Seasonal shifts in age composition were also reported whereby adult loggerheads were dominant during spring and summer months (males: April-May; females: May-August) and juveniles more abundant during the rest of the year. Juvenile arrival back into the area coincided with the departure of adults during late summer and juvenile departure coincided with arrival of breeding adults in the spring. Furthermore, Carr et al. (1980) confirmed the presence of overwintering loggerheads in both the juvenile and maturing size classes. In that study, torpid turtles were trawl collected from the channel during February and were believed to have been at least partially imbedded in muddy bottom sediment. Eighty-five percent of the turtles captured were reported as juvenile or sub-adult (i.e. close to maturing size) individuals. Schroeder and Thompson (1987) also confirmed the presence of loggerheads during winter months by way of aerial surveys. In that study, turtles were observed in both inner and mid-shelf habitats and the authors suggested the majority of these were juvenile or maturing size animals.

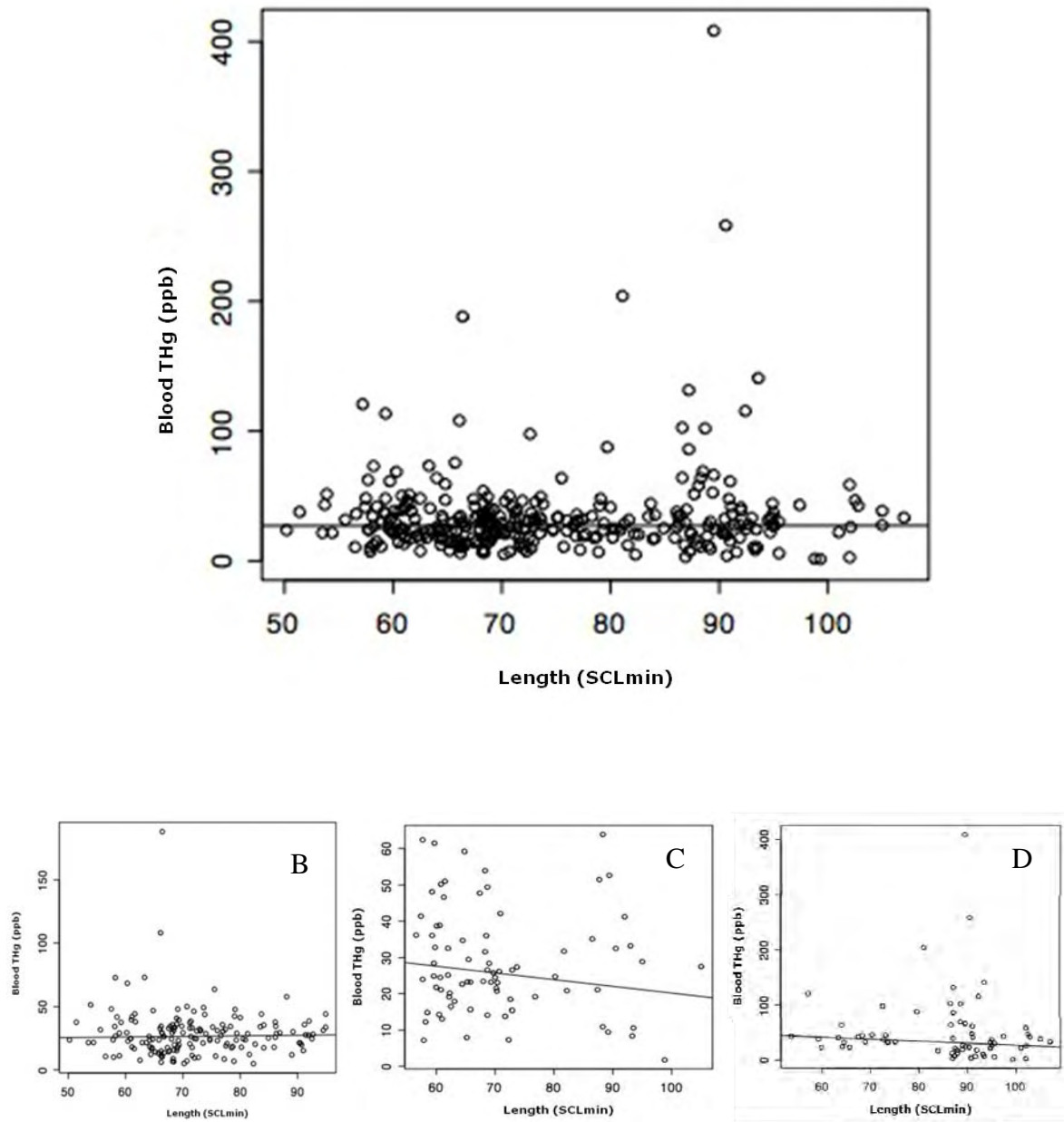
SCDNR sampling targeted adult turtles in the Port Canaveral channel. However, 67% ( $n=107$ ) of the turtles captured in this location were juveniles. Subsequent Hg analysis included 68 Port Canaveral turtles, 18 of which were juveniles. Blood THg among Port Canaveral juveniles was significantly different than juveniles from all other locations (K-W,  $H=65.669$ ,  $p=3.61E-14$ ). Recapture of juvenile turtles tagged in the Port Canaveral area suggests northward movement of juveniles to foraging habitats off the Carolinas and mid-Atlantic U.S. during the summer months (Henwood, 1987b; Schmid, 1995; Morreale and Standora, 2005). Morreale and Standora (2005) also summarized evidence of the southerly fall migration of loggerheads from northern foraging habitats subject to suboptimal winter temperatures. Satellite tracking evidence of a return to waters off Port Canaveral during the winter is not currently available. However, given the evidence of significant numbers of overwintering juveniles, either within the channel or waters further out on the coastal shelf, a winter return for at least some individuals seems plausible. Satellite tracking data collected thus far suggests juvenile loggerheads migrate from northern foraging habitats to points at least as far as North Carolina (Morreale and Standora, 2005). However, Morreale and Standora (2005) indicated at least some juvenile loggerheads may continue on or ultimately be forced as far south as Cape Canaveral where winter water temperatures would consistently be sufficient to ride out the winter months. Carr et al. (1980) indicated overwintering turtles within the Port Canaveral channel were not believed to be foraging. However, if juvenile turtles do return to warmer midshelf waters off Port Canaveral and continue foraging during winter, sustained Hg exposure could be one consequence. Interestingly, SCDNR sampling would most likely miss the capture of many juveniles overwintering in the Port Canaveral area. Most of these turtles may be expected to return to waters north of the SCDNR sampling area in the spring and their departure should occur earlier than any SCDNR sampling would begin in this area, given the timeline established by Henwood (1987b) and Schmid (1995). Subsequent summer sampling in estuarine and near shore waters from North Carolina northward (possibly as far as New York) may be required to examine the effect on blood Hg due to overwintering in the Port Canaveral area.

Overall, blood THg was not significantly different between adults from Port Canaveral and adults from all other locations (K-W,  $H=0.843$ ,  $p=0.655$ ; north Florida omitted due to small sample size). This is most likely due to the large standard deviation among Port Canaveral adults. Both Henwood (1987b) and Schmid (1995) reported the general departure of adults from the Port Canaveral channel upon conclusion of breeding and nesting. Adult males and females were then believed to take different migratory routes to foraging habitats. However, other studies have indicated the presence of adult size turtles in these waters during winter months. The collection of tagged males in successive years led Schmid (1995) to conclude at least a portion of the adult males at Port Canaveral may overwinter in the area. Satellite tracking data from the adult males tagged by SCDNR indicated at least some individuals remain in coastal waters off Port Canaveral for significant periods of time after the breeding season (Arendt et al., 2007; Chapter 6 of this report). Satellite data for two of these turtles indicated their presence in Port Canaveral near shore waters as late as mid-November. Similar concentrations between both transient/resident satellite-tagged males and juvenile/adult turtles were observed. The lack of differences between these groups suggests juveniles and adults, although highly mobile at different times of the year, may both spend significant amounts of time in waters off Port Canaveral, thereby integrating the Hg exposure they receive at this location. These data also suggest adults may remain offshore of Port Canaveral longer than originally thought upon

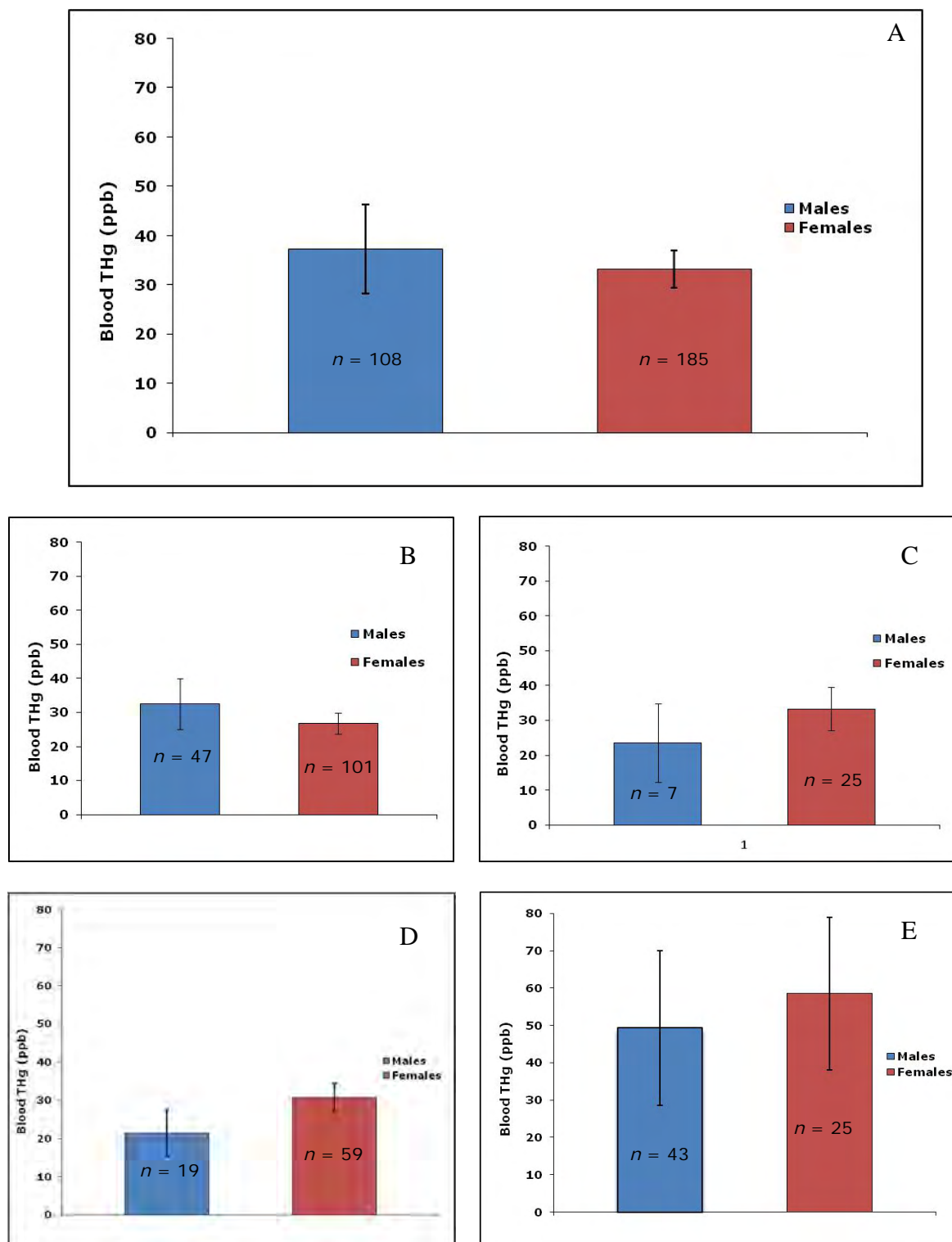
conclusion of breeding. This region has long been recognized for its importance to global loggerhead nesting, representing the highest nesting concentration in the western hemisphere (Ehrhart and Raymond, 1983; Schroeder and Thompson, 1987) and one of the two most significant loggerhead nesting aggregations worldwide (Ehrhart and Raymond, 1983; Ehrhart et al., 2003, Witherington et al., 2009). Further sampling of adults for Hg studies should be a goal of continued research in the Port Canaveral area, given the small sampled sizes for adult male ( $n=43$ ) and female ( $n=25$ ) loggerheads at this location. Any potential link between loggerhead nesting decline (Witherington et al., 2009) and anthropogenic contaminants in this area should definitely be investigated.

Lastly, we should note the data presented here are not meant to imply any overt health risk to loggerheads in southeastern U.S. waters. The data suggest overall Hg concentrations in the southeast loggerhead population are rather low. Still, caution must be exercised when interpreting the results. This study described Hg concentrations in the blood compartment only; however, the blood compartment is intricately linked to internal tissues where high Hg concentrations have been shown to have detrimental effects. Moreover, blood is the pathway by which Hg ingested with the diet is distributed to other tissues. Day (2003) and Day et al. (2005) reported blood THg was significantly related to THg concentrations in several internal tissues in stranded loggerheads from the southeastern U.S., including: muscle, spinal cord, kidney, and liver ( $R^2 > 0.89$  for each relationship). The strongest of these relationships were between blood and muscle or spinal cord. In the absence of Hg data from internal tissues, the relationships presented in those papers may provide a rough estimate of internal Hg burdens for loggerheads in the current study. The mean blood THg concentration of all turtles in this study was 36.13ppb. This concentration would result in relatively low muscle and kidney THg concentrations of approximately 67ppb and 26ppb respectively, given the regression equations reported by Day et al. (2005). However, a blood THg concentration of approximately 40ppb might result in a liver THg concentration near 400ppb (0.4 parts per million, ppm) and a kidney concentration near 150ppb (0.15ppm) given relationships for these compartments in stranded loggerheads presented by Day (2003). For comparison, the U.S. Food and Drug Administration and the U.S. Environmental Protection Agency have established fish advisory levels between 0.3 and 1.0ppm depending on the consumer.

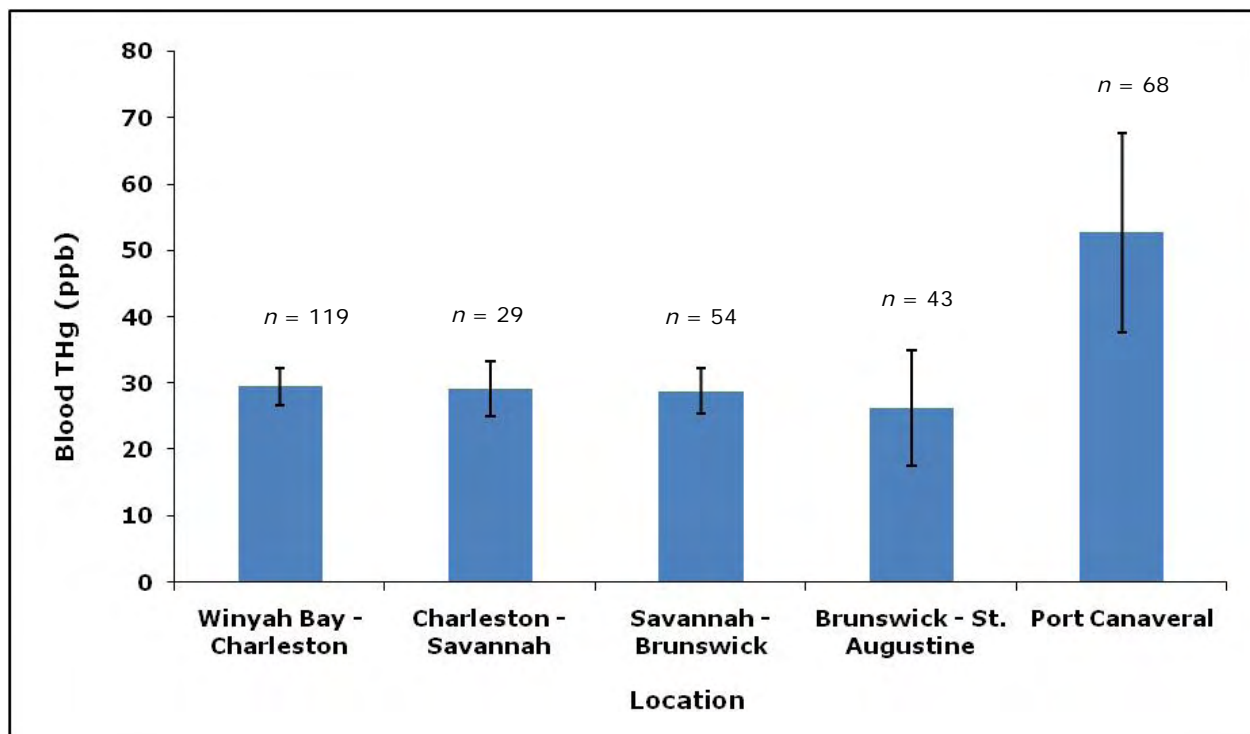
Although the blood concentrations presented here may not be sufficiently high to elicit overt health effects; however, blood THg concentrations for loggerheads sampled in 2003 and reanalyzed in this report have been shown to compromise immune response (Day et al., 2007). *In vitro* exposure to methylmercury concentrations consistent with the level of blood THg concentrations of approximately 5% of the 2003 loggerheads (since the majority of blood THg has been shown to be in the methylated form) resulted in immunosuppression, including effects on B and T cell response. The maximum THg concentration for all 2003 loggerheads was 77ppb (Day et al., 2007), less than 1/5 the concentration of the highest blood THg concentration recorded among Port Canaveral turtles. Additionally, the range of THg concentrations seen among turtles analyzed by Day et al. (2007) would include roughly 95% of all turtles analyzed for Hg in the current study. Thus, further examination of relationships between THg and clinical blood parameters or immune response may reveal similar negative impacts for turtles sampled during the various studies covered in this report.



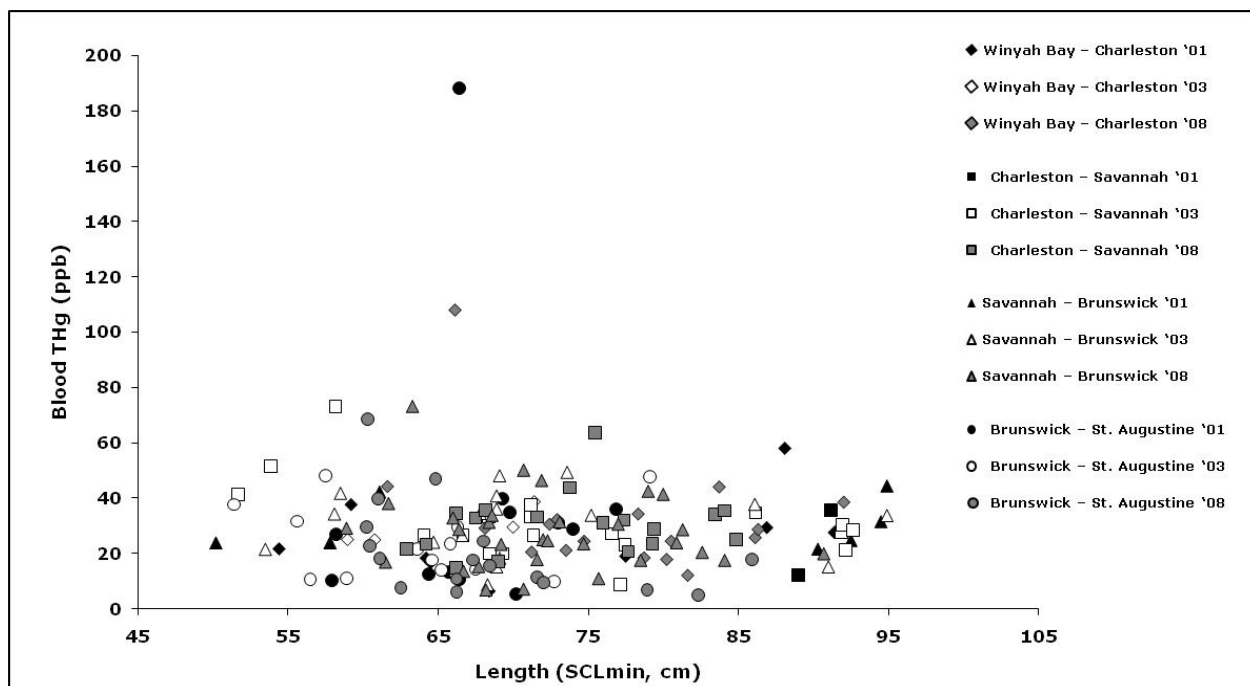
**Figure 6.1.** Kendall's regression between blood THg (ppb) and loggerhead length (SCLmin,cm). A) Complete data set 2001-2008. B) Regional index of abundance study 2001, 2003, 2008. C) Charleston, SC ship channel. D) Port Canaveral, FL ship channel.



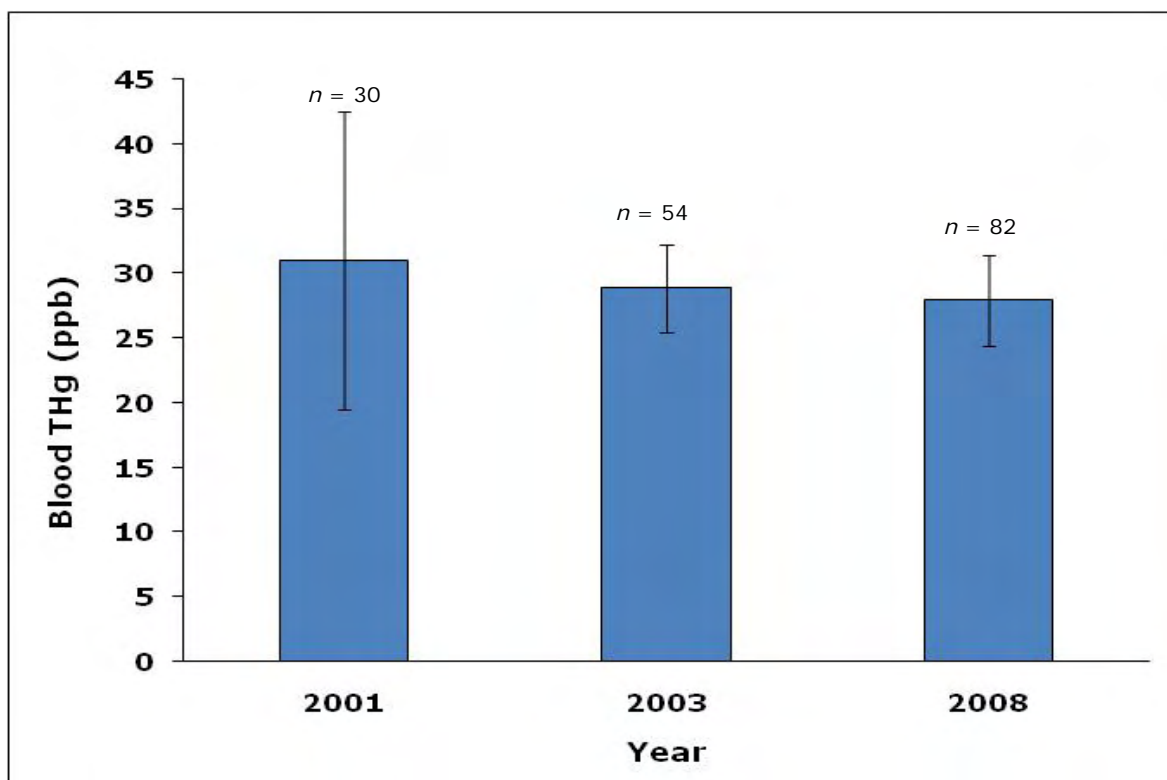
**Figure 6.2.** Mean blood THg concentration ( $\pm$  95% CI) between sexes. A) Complete data set 2001-2008. B) Regional index of abundance study 2001, 2003, 2008. C) Charleston, SC ship channel satellite tagged loggerheads. D) All Charleston, SC ship channel loggerheads. E) All Port Canaveral, FL ship channel loggerheads.



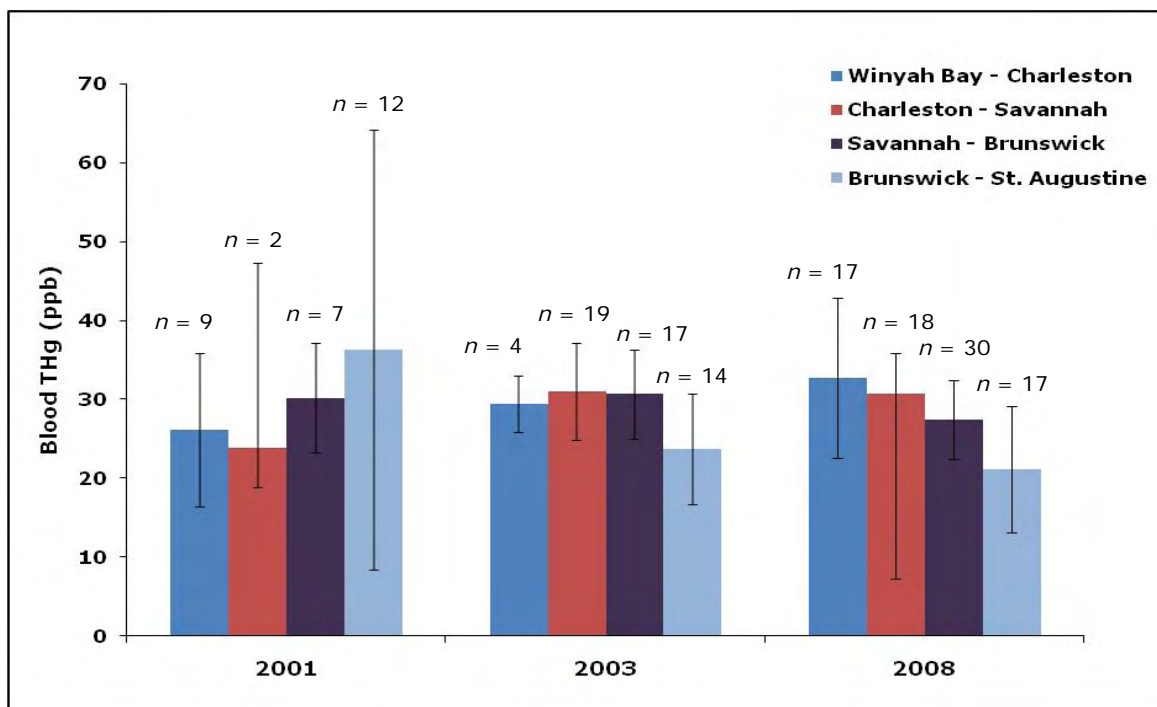
**Figure 6.3.** Mean blood THg concentration ( $\pm$  95% CI) for loggerheads captured within four sub-regions from South Carolina to Florida during SCDNR sampling 2001, 2003-2008.



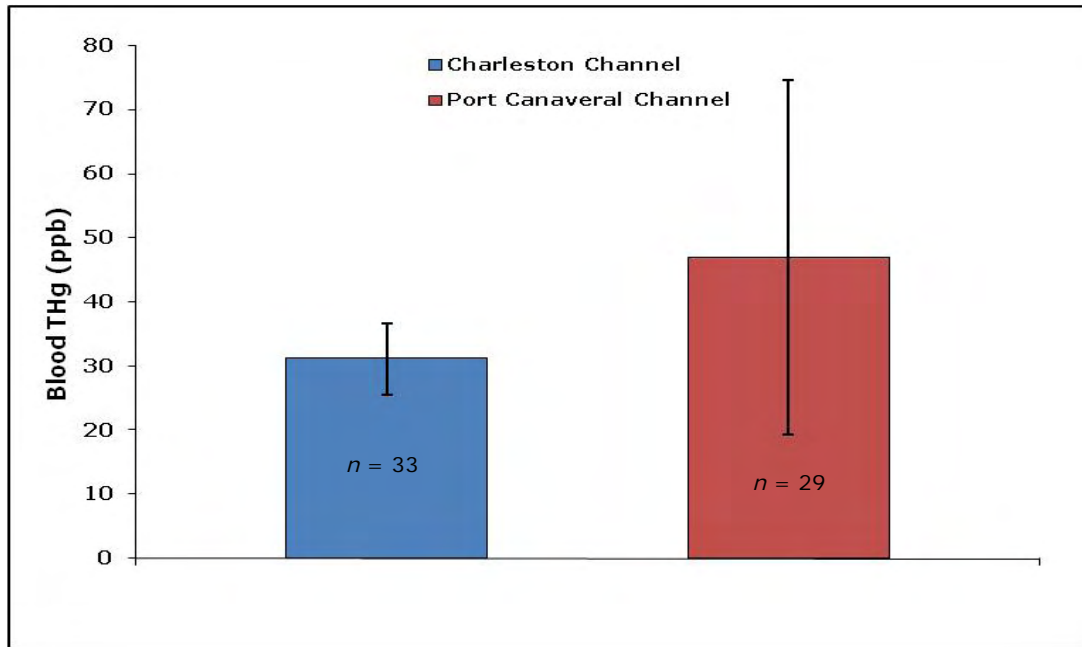
**Figure 6.4.** Scatter plot of blood THg versus SCLmin for all loggerheads captured within four sub-regions from South Carolina to Florida during the regional index of abundance study (2001, 2003, 2008).



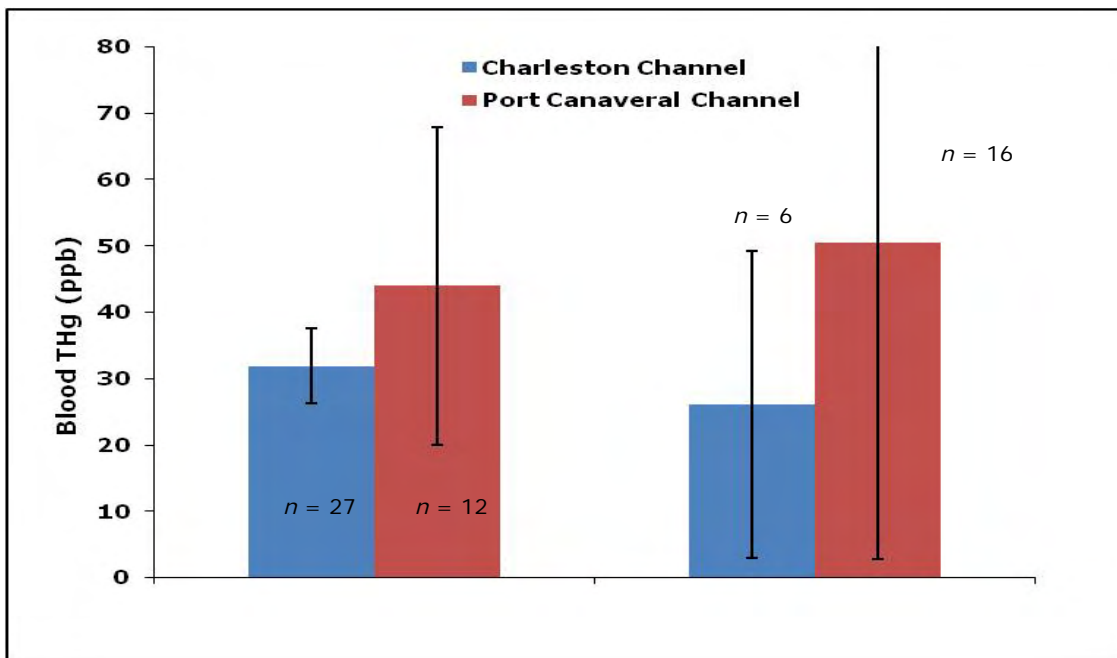
**Figure 6.5.** Mean blood THg ( $\pm$  95% CI) between years for all loggerheads captured during the regional index of abundance study.



**Figure 6.6.** Mean blood THg ( $\pm$  95% CI) for loggerheads captured within four sub-regions from South Carolina to Florida during the regional index of abundance study (2001, 2003, 2008).



**Figure 6.7.** Mean blood THg ( $\pm$  95% CI) for loggerheads fitted with satellite tags during targeted sampling in the Charleston, SC and Port Canaveral, FL shipping channels.



**Figure 6.8.** Mean blood THg ( $\pm$  95% CI) of resident and transient loggerheads fitted with satellite tags in the Charleston, SC and Port Canaveral, FL shipping channels. In general, “resident” Charleston-tagged loggerheads remained within 40km off the coast of SC, GA and nFL. “Transients” rapidly emigrated out of the regional trawl survey area (2000-2003) and did not return. Port Canaveral “residents” generally remained in near shore waters in the vicinity of the Port Canaveral channel. Between mid-May and early June, transients rapidly dispersed to distant locations both north and south of the state of Florida (see Chapter 5).



## **Chapter 7 By-catch assessment and relationships between by-catch and turtle occurrence in trawl surveys in coastal waters of South Carolina, Georgia, and northern Florida.**

### **Introduction**

To manage and protect marine sea turtle species, scientists must be able to characterize their movements and behaviors on both a large scale and on a local level. There are a number of factors that could influence movements and the occurrence of sea turtles on a local scale, including habitat preferences and the availability, abundance, and distribution of prey items. Along the Atlantic coast of the United States, loggerhead (*Caretta caretta*) sea turtles occupy a diversity of near shore habitats (Hopkins-Murphy, et al. 2003). Large immature and adult turtles tend to occur in association with both natural and manmade hard substrates (Hopkins-Murphy et al. 2003). Lohofener et al. (1989) found that turtles were significantly associated with petroleum production structures offshore of the Chandeleur Islands in the Gulf of Mexico, but this association was not as clear in other offshore study areas to the west where turtles were less abundant. Grays Reef off the Georgia coast, live bottom areas off east-central Florida, and The Flower Gardens coral reef 150km off the Louisiana/Texas border are known turtle resting grounds where turtles seem to be present the majority of the time (Hickerson and Peccorini, 2000; Hopkins-Murphy et al., 2003). Although it is not understood how loggerheads use structured, hard bottom habitats, or whether they favor them over softer substrates, it is thought that they prefer habitats with hard structures (Hopkins-Murphy et al., 2003).

The feeding habits and diet of loggerheads have been widely reviewed (Mortimer, 1982; Lutcavage and Musick, 1985; Plotkin et al., 1993; Burke et al., 1993; Youngkin and Wyneken, 2005; Seney and Musick, 2007). Loggerheads are carnivorous, feeding primarily on benthic invertebrates (Plotkin et al., 1993). Throughout their geographic distribution they forage on a diversity of prey items (Plotkin et al., 1993) and have been shown to shift their diet seasonally (Plotkin et al., 1993; Youngkin and Wyneken, 2005) and temporally (Youngkin and Wyneken, 2005; Seney and Musick, 2007), perhaps due to specific prey items becoming more available and/or abundant. Studies on stranded loggerheads in Virginia (Seney and Musick, 2007), Georgia (Youngkin and Wyneken, 2005), and Texas (Plotkin et al., 1993) have shown that crabs, mollusks, and finfish make up a substantial proportion of a loggerhead's diet. However, it is thought that loggerheads cannot swim or maneuver fast enough to catch finfish species (Youngkin and Wyneken, 2005). The finfish species found in stranded loggerhead guts were also the predominant species observed in trawl by-catch, perhaps indicating that turtles with finfish in their gut were feeding on by-catch from shrimp trawls (Youngkin and Wyneken, 2005).

To better understand some of the factors that could affect loggerhead occurrence, by-catch from randomized sea turtle trawl surveys in SC, GA, and northern FL was assessed. By-catch data were recorded during the 2008 regional sea turtle trawl survey and by-catch data from all five years (2000-2003, 2008) of the regional survey were analyzed to determine if any differences were observed in the frequency of occurrence of sea turtle prey species. Additionally, data were analyzed to determine if relationships existed between the occurrence of potential prey species or bottom habitat and the occurrence of loggerhead turtles.

## Methods

### *Data Collection*

Trawling was conducted at randomly selected locations in coastal waters (4.6 to 12.2m deep) corresponding to the shoreline between Winyah Bay, SC, and St. Augustine, FL. Sampling was conducted between May and August 2000, and May through July in 2001-2003 and 2008. During 2008, additional cruises were conducted in August at turtle “hot spot” locations; however, these trawl events were excluded from the data set because they were not randomly selected and many stations were duplicated multiple times throughout each cruise.

At each station a tow was made with two 19.8m (65ft) flat trawls consisting of 20.3cm (8in) stretch mesh webbing in the body and 10.2cm (4 in) stretch mesh in the tailbag. During 2000-2003, thirty-minute tows (bottom time) were made at each station. In 2008, tows were reduced to twenty-minutes (bottom time) due to changes in the project’s Federal permit. Additionally, a 4.6m (15ft) try-net with 1.9cm (0.75in) stretch mesh was also fished to obtain abundance on species co-occurring with sea turtles during 2000-2003. This net was towed for 15 minutes bottom time during the first part of turtle trawl tows. By-catch data collected from the try-net were excluded from all analyses except those done to characterize bottom habitat.

By-catch collected from the port and starboard nets were sorted separately; elasmobranchs, finfish and invertebrates were counted and identified to the lowest practical taxonomic level. For some trawl events, data recorded represent estimates of by-catch rather than true counts. Estimates were taken for the following types of collections: those containing multiple turtles (due to time constraints and priority of processing turtles first); those containing exclusively jellyfish or those with animals too large to be brought on board (if possible, individual counts were made prior to dumping net contents over the side of the vessel); and those with large numbers of by-catch items caught in the wings of the nets. When possible, collective weights (to the nearest kg) were obtained for non-priority finfish and non-priority invertebrates.

Priority organisms included elasmobranchs, commercially and recreationally important finfish and invertebrates, and sea turtle prey items. For each elasmobranch, sex and total length (to the nearest cm) were recorded prior to release. Total length and collective weight of each species was recorded for priority finfish. For priority invertebrates (with the exception of cannonball jellyfish, *Stomolophus meleagris*), total length or carapace width of individuals was measured to the nearest centimeter and each species was weighed collectively.

### *Data Analysis: By-catch Assessment*

Species number ( $s$ ) for each trawl event was calculated by removing undetermined species. In the case of species for which more than one level of identification was included, the more general classification was excluded (i.e. if *Prionotus* sp. and *Prionotus carolinus* were both present, *Prionotus* sp. would be excluded). All species classifications were used in the calculation of abundance (number of individuals;  $n$ ). By-catch groups (elasmobranchs, finfish, and invertebrates) were divided into 16 sub-groups (see Figure 7.1) for descriptive analyses.

Sampling stations were divided into three groups: stations located in strata 27-36 (St. Augustine, FL to St. Catherine’s Inlet, GA), strata 37-42 (St. Catherine’s Inlet, GA to St. Helena Sound,

SC), and strata 43-50 (St. Helena Sound, SC to Winyah Bay, SC). These groups were designated as the southern, central, and northern sub-regions, respectively, based on regions delineated in Maier et al. (2004). Frequency of occurrence (% of trawls with species present vs. absent) was used in lieu of relative abundance because estimates instead of counts were often reported. Chi-square analyses were used to evaluate regional differences in the frequency of occurrence of major by-catch groups and sub-groups across sub-regions in 2008.

Temporal differences were analyzed for selected species (Appendices A-C). For elasmobranchs, CPUE was deemed an appropriate representation of catch since most data for this group represented counts rather than estimates. In addition, size-selectivity of trawl mesh should not have excluded elasmobranchs. Standardized CPUE (organisms per 30.5m net-hr) was calculated by the method described in Jamir (1999) and detailed in Chapter 2. CPUE data could not be normalized through transformation; thus, data were analyzed using the nonparametric Kruskal-Wallis tests; Mann-Whitney U-tests were also used as non-parametric post-hoc tests to evaluate inter-annual differences). Non-parametric tests examine differences among ranks; however, CPUE graphs in this section depict means. For finfish, frequency of occurrence was analyzed across study years (using chi-square tests) in lieu of CPUE, for reasons previously stated.

Potential prey species were divided into nine groupings for analyses: blue crab (*Callinectes sapidus*), Portunid crab (*Ovalipes*, *Portunus*, *Arenaeus*, *Callinectes similis* and *C. ornatus*), spider crab (*Libinia sp.*), stone crab (*Menippe mercenaria*), miscellaneous crab, whelk/conch (*Busycon sp.* and *Pleuroploca gigantia*), horseshoe crab (*Limulus polyphemus*), cannonball jellyfish (*Stomolophus meleagris*), and miscellaneous jellyfish (to include Ctenophora). Frequency of occurrence (percentage of trawls with species present vs. species absent) was used to assess temporal and regional trends for potential prey species. Frequency of occurrence was used in lieu of relative abundance (CPUE) for reasons stated above. Chi-square analyses were used to determine if regional or temporal differences existed in the selected prey species group.

#### *Data Analysis: By-catch Species and Turtle Occurrence*

Data collected in 2000-2003 and 2008 were used to evaluate relationships between loggerhead occurrence and bottom habitat and loggerhead occurrence and known prey species. For this analysis, by-catch species were used to characterize bottom habitat based on methodologies developed in Van Dolah et al. (1994). Each trawl event [consisting of port, starboard, and try-net when available (2000-2003)] was classified as either hard bottom habitat, probable hard bottom habitat, or not hard bottom habitat based on the co-occurrence of three or more, two, or less than two reef indicator species, respectively. Finfish indicator species were identified in Van Dolah et al. (1994), and sponge, coral, and bryozoans were identified as invertebrate indicator species by Reed (2004). Logistic regression was used to determine if relationships existed between the occurrence of loggerheads and bottom type and the nine prey species groups. Regional comparisons using the previously mentioned sub-regions were also performed.

Data analyses utilized R statistical software (R Development Core Team, Vienna, Austria; <http://www.r-project.org>) or Statistical Package for the Social Sciences (SPSS Version 13.0).

## Results

### 2008 Overview

By-catch collected in 2008 consisted of 87,753 individuals comprising at least 142 different species (18 species of elasmobranchs, 61 species of finfish and 63 species of invertebrates). Finfish were the numerically dominant group with 50,698 individuals, followed by invertebrates with 35,461 individuals. Elasmobranchs were the least abundant with 1,594 individuals.

Midwater/pelagic finfish was the most abundant subgroup in 2008 (Figure 7.1) due to high catches of Atlantic bumper (*Chloroscombrus chrysurus*; 28,972 individuals) and Atlantic moonfish (*Selene setapinnis*; 7,939). Invertebrate catches were dominated by jellyfish due to high catches of sea nettles (*Chrysaora quinquecirrha*; 11,194) and cannonball jellies (*Stomolophus meleagris*; 6,851). Another numerically dominant invertebrate sub-group was the echinoderms, with consistently large catches of gray sea stars (*Luidia clathrata*; 3,717) and short-spined sea urchins (*Lytechinus variegatus*; 2,946).

Number of by-catch species in 2008 was highest in the northern sub-region (115 taxa) and lowest in the central (101 taxa). The number of finfish species numbers was highest in the southern and lowest in the central sub-regions (Figure 7.2). Elasmobranch and invertebrate species decreased slightly from north to south.

Elasmobranchs occurred in approximately 65% of trawls during summer 2008. Chi-square analyses found regional differences for the elasmobranch group in 2008 (Table 7.1). Occurrence of elasmobranchs was highest in the northern sub-region (73%) followed by the central (67%) and southern (57%) sub-regions, respectively. The only subgroup with significant regional differences within the elasmobranch group was sharks (Table 7.2), which displayed the same pattern of decreasing frequency of occurrence in trawls from north (64%) to south (47%). The sub-groups skates and rays were not analyzed, as the number of trawls with skates and rays in each sub-region in 2008 was too small for chi-square analysis.

Finfish frequency of occurrence was not significantly different among sub-regions in 2008 (Table 7.1), although there were regional differences in all finfish subgroups except midwater/pelagic fishes (Table 7.3). Demersal fishes and flatfishes occurred with the highest frequency in the central sub-region (62% and 25.5%, respectively) and the lowest frequency in the southern (49% and 17%, respectively). Inshore fishes occurred most frequently in the northern sub-region (34%) and least frequently in the south (22%). Occurrence of reef fishes was greatest in the central sub-region (75%) and least in the north (60%).

Frequency of occurrence of invertebrates was not significant among regions in 2008 (Table 7.1), although there were regional differences in the subgroups crabs/shrimp, jellyfish, and sessile invertebrates (Table 7.4). Crabs/shrimp and sessile invertebrates were found most frequently in trawls in the central sub-region (60% and 74%, respectively) and least frequently in the southern (44% and 52%, respectively). Jellyfish occurred most frequently in the southern sub-region and decreased from south (64%) to north (52%). Horseshoe crabs were not analyzed, as the number of trawls with horseshoe crabs in one of the sub-regions was too small for chi-square analysis.

### *Temporal Comparison*

Overall species numbers (sub-regions combined) increased over the study period, with approximately 16% more taxa recorded in 2008 than in 2000 (142 vs. 122; Figure 7.3). Elasmobranch species numbers were generally consistent throughout the study; however, 2.75 fewer taxa were collected in 2008 than in previous study years. Finfish species numbers have increased by 45% since 2000 (although there was a decrease from 71 in 2003 to 61 in 2008). Numbers of invertebrate species appeared to decrease steadily over the years until a 62% increase between 2003 and 2008. This dramatic rise in invertebrate species may have been an artifact of variations in classification schemes used to identify organisms each year, as well as intensified efforts in 2008 to properly identify and quantify invertebrate by-catch. Additional comparisons for selected priority species, including CPUE or frequency of occurrence, as well as size ranges, mean sizes, and/or sex ratios are found in Appendices A-C.

### *Occurrence of Turtle Prey Items*

Prey species occurred in over 50% of all trawls in all three sub-regions during all five years of the regional survey. Overall occurrence of prey species was highest in the central sub-region (84.9%) followed by the southern (78.6%) and northern (69.8%) sub-regions. Lowest frequencies of prey occurrence (between 51-56%) were found in the northern sub-region in 2002 and in the southern sub-region during 2003. Frequencies of prey occurrence in at least 90% of all trawls were found in the central sub-region in 2001 and in the southern sub-region in 2001 and 2002. In 2008, occurrence of prey species was similar among all regions (Table 7.5). Initial analyses found temporal differences for all nine prey groups and regional differences for all prey groups, except blue crab and horseshoe crab. Since blue and horseshoe crab displayed no regional differences, all regions were grouped together for further analyses. All other prey groups were divided into sub-regions, with the exception of whelk/conch. The number of trawls with whelk/conch in each sub-region was too small to allow further analysis. Figures 7.4 – 7.12 show frequency of occurrence for all prey groups throughout the project for each sub-region.

The frequencies of trawls with cannonball jellyfish and miscellaneous jellyfish were the highest occurrences for any of the prey groups across all three sub-regions. Significant temporal differences were found in the northern ( $p < 0.001$ ), central ( $p < 0.001$ ), and southern ( $p < 0.001$ ) sub-regions for cannonball jellyfish. Frequency of trawls containing cannonball jellyfish decreased in the northern sub-region from 2000-2003 with an increase observed in 2008; the highest frequency observed for any one prey species for the northern sub-region throughout the project. Frequencies in the central sub-region were highest during 2001-2003. In the southern sub-region, cannonball jellyfish occurred in low frequency during 2000 with a pronounced increase by over 60% in 2001. These frequencies subsequently decreased in the southern sub-region from 2001-2003 with another increase in frequency observed in 2008. Significant temporal differences were also noted for miscellaneous jellyfish in all three sub-regions (northern  $p < 0.001$ ; central  $p = 0.001$ ; southern  $p < 0.001$ ). The lowest occurrence of miscellaneous jellyfish was observed in the northern sub-region in 2000 and the highest frequencies were observed in the southern sub-region in 2002. In 2003, frequency of occurrence of miscellaneous jellyfish in the southern region decreased by over 55%.

No significant regional differences were observed in the frequency of occurrence of blue crab in trawl events; however, temporal differences were significant ( $p < 0.001$ ). The frequency of trawls with blue crab present was highest from 2000-2001. In 2002, there was a drop in frequency that corresponded with low catch rates in a directed blue crab survey (SCDNR, 2004) which was attributed to a severe 5-year drought that peaked in the southeast US in summer 2002. Rainfall increased in the fall of 2002 through spring 2003, ending drought conditions (SCDNR, 2003) and in turn improving environmental conditions for blue crab. However, only slight increases in the frequency of trawls with blue crab were seen from 2003 to 2008 in our survey, perhaps due to the return of severe drought conditions to the southeastern US during 2008.

Significant temporal differences were found in all three sub-regions for Portunid crab (northern  $p = 0.002$ ; central  $p < 0.001$ ; southern  $p < 0.001$ ), spider crab (northern  $p < 0.001$ ; central  $p = 0.029$ ; southern  $p < 0.001$ ), and miscellaneous crab (northern  $p < 0.001$ ; central  $p < 0.001$ ; southern  $p < 0.001$ ). A large decrease in frequency of occurrence of Portunid crab was observed in the central and southern sub-regions in 2003; however, frequencies in all sub-regions increased from 2003 to 2008. Increases in the frequency of occurrence of spider crabs were observed from 2000 to 2001 in all sub-regions with the largest increase in the southern sub-region. Decreases in frequency of spider crab occurred in all three sub-regions from 2001-2003. In 2008, spider crab frequencies increased in the northern and southern sub-region (consistent with cannonball jellyfish, which spider crabs use for benthic-pelagic transport) while frequencies in the central sub-region remained stable. Highest frequency of occurrence of miscellaneous crabs was observed in the central sub-region.

No significant regional differences were found in horseshoe crab frequencies; however, temporal differences were significant ( $p < 0.001$ ). The frequency of trawls with horseshoe crabs present remained relatively stable from 2000-2003, with a large increase in frequency, particularly in the northern and southern sub-regions, observed during 2008. Significant temporal differences were found in the frequency of occurrence of stone crabs in the central ( $p < 0.001$ ), and southern ( $p < 0.001$ ) sub-regions. Frequency of occurrence of stone crabs remained relatively stable over the study period within each sub-region with the exception of large increases observed in the central and southern sub-regions in 2001. The frequency of trawls with whelk/conch present was the lowest occurrence for any one prey species across all three sub-regions.

#### *By-catch Species and Turtle Occurrence*

Sampling sites classified as hard bottom habitat, probable hard bottom habitat, and not hard bottom habitat were found in all five years of the regional survey and among all three regions within each year. Pearson Chi-square showed that the relative proportion of bottom habitat types were significantly different among years ( $p = 0.01$ ) with the largest proportion of hard bottom habitat found in 2000 and the smallest proportion found in 2002 (Table 7.6). Differences among years could be attributed to variation in sampling sites due to random selection or patchiness of hard bottom throughout the sampling region, but could also be influenced by differences in the crew's ability to identify by-catch species. During 2008, 2.9% of all sites sampled were classified as hard bottom habitat and 8.1% were classified as probable hard bottom habitat. These numbers are comparable to the mean percentage of sites identified as hard bottom habitat (2.8%) and potential hard bottom habitat (7.9%) over the five years of the regional survey. The

relative proportion of bottom habitat types was also significantly different among regions ( $p=0.04$ ) with the highest percentage of hard bottom habitat found in the northern region. In four of the five years of the regional survey, the northern region contained the most sites classified as hard bottom. However, during 2008, the central region was found to have the highest percentage of both hard bottom (4.3%) and probable hard bottom habitat (17.3%; Table 7.6).

Previous analysis of by-catch data collected from the 2000-2003 in-water regional survey utilized correlation, Chi square and cluster analyses to determine if relationships existed between the presence of loggerhead turtles and individual by-catch species or groups of by-catch species (Byrd et al., 2008). These analyses found no significant relationships between any of the by-catch species and the presence of loggerhead turtles (SCDNR, unpublished data). Logistic regression analyses were used to determine if relationships existed between the occurrence of loggerhead turtles and a specific bottom habitat or a specific prey group. Initial analyses were run to determine if temporal and regional differences existed with the occurrence of loggerhead turtles and bottom habitat. No significant temporal differences were seen with the occurrence of loggerhead turtles ( $p = 0.992$ ). However, significant differences were detected between regions ( $p < 0.001$ ; Table 7.7), warranting further analysis on a regional level.

In the northern sub-region, no significant relationships were observed between the occurrence of loggerheads and bottom type for eight of the nine prey groups (Table 7.8). A significant inverse relationship was seen between the presence of spider crabs and the presence of loggerhead turtles. Trawls were 0.490 times more likely to catch a loggerhead when spider crabs were not present. In the central sub-region, no significant relationships were seen between the occurrence of loggerheads and bottom type for seven of the nine prey groups (Table 7.9). Significant inverse relationships were seen between the presence of spider crab and miscellaneous jellyfish and the presence of loggerhead turtles. Trawls were 0.598 times more likely to catch a loggerhead when spider crabs were not present and were 0.569 more likely when miscellaneous jellyfish were not present. In the southern sub-region, no significant relationships were seen between the loggerhead occurrence and bottom type for eight of nine prey groups (Table 7.10). A significant inverse relationship was seen between the presence of spider crabs and the presence of loggerhead turtles. Trawls were 0.384 times more likely to catch a loggerhead when spider crabs were not present. Although the inverse relationships between the presence of spider crabs and the presence of loggerheads in all three sub-regions and the presence of miscellaneous jellyfish and loggerheads in the central region were statistically significant, they may simply be an artifact of the data set instead of meaningful biological relationships. However, it is interesting to note that in all three sub-regions the analysis found that the presence of spider crabs, a preferred loggerhead prey item, was inversely related to the presence of loggerheads.

## **Discussion**

Analysis of by-catch could potentially give insight into the probability of occurrence or catch rates of sea turtles in trawl surveys. A variety of environmental factors, including water temperature and rainfall, will influence the abundance and availability of prey species within the project's sampling area. Although regional and temporal differences were seen in almost all of the prey groups analyzed, there were multiple prey species available, albeit in different frequencies, throughout all three sub-regions in all five years of the regional survey. Diet studies

have found that loggerheads eat a variety of prey items (Plotkin et al., 1993; Youngkin and Wyneken, 2005; Seney and Musick, 2007) and will opportunistically eat whatever prey is available (Plotkin et al., 1993; Seney and Musick, 2007). Since a variety of prey items were available throughout all years of the regional survey, differences seen in catch rates of turtles may not be due to differences in frequencies of particular prey items. However, it is important to note that the analysis in this chapter investigated differences in frequency of occurrence of prey groups and did not look at relative abundance or prey densities. In the future, the historic Southeast Area Monitoring and Assessment Program South Atlantic (SEAMAP-SA) trawl data set could be used to investigate trends in prey densities over time and the potential relationship of these prey densities with turtle occurrence and catch rates.

Stations classified as hard bottom habitat and known loggerhead prey items were found in all three sub-regions throughout the entire study period. Although loggerheads are thought to prefer hard bottom habitat (Hopkins-Murphy et al., 2003) and have been found to pursue mobile prey (Plotkin et al., 1993), no strong relationships were found between the presence of loggerhead turtles with a particular bottom habitat and few significant relationships were found between the presence of loggerhead turtles and prey groups. Spider crabs were the only prey species found to have a significant relationship with the occurrence of loggerhead turtles in all three sub-regions and this relationship was actually negative despite spider crabs being a known prey item (Plotkin et al., 2003; Youngkin and Wyneken, 2005; Seney and Musick, 2007). This relationship may actually be an artifact of the dataset rather than a meaningful biological relationship.

Although analyses did not show strong relationships between the occurrence of loggerhead turtles and bottom habitat or known prey items, it does not necessarily mean that these relationships do not exist. Sampling methodologies used in this study may be a factor in why these biological relationships were not seen. The trawl gear used in this study ineffectively samples hard bottom. Less than 5% of all of the sampling sites were classified as hard bottom using the technique described in the methods section. Under-sampling hard bottom habitats helps to avoid gear loss and habitat destruction, however, it also likely reduces the potential for any relationships to be seen between turtle occurrence and habitat type. Furthermore, since the primary objective of the study was to catch turtles, the trawl nets used for sampling were constructed of large mesh to limit the amount of by-catch caught and potentially harmed. Due to this large mesh size, many of the potential prey and reef indicator species may be able to pass through the nets undetected. Therefore the catch that is brought onto the boat for work-up may not accurately represent all the species that occurred in the area trawled. It is also important to note that this analysis relies on the presence or absence of species recorded for each trawl event. It is likely that some potential reef indicator species or known prey items may not have been counted or recorded for a number of events, particularly during events when multiple turtles were caught. In many of these situations, by-catch was dumped overboard in order to concentrate crew efforts on processing the turtles. If by-catch species were not accurately recorded for sampling events, particularly events where turtles were caught, it would influence the likelihood of finding relationships between turtles and individual prey species.



**Table 7.1.** Summary of statistical analyses of frequency of occurrence by sub-region for major by-catch groups in 2008, showing both significant and non-significant (NS) differences and associated p-values. Determination of significance is based on  $p < 0.05$ .

2008 Mean Abundance ( <i>n</i> ) by Sub-region - Groups			
	<b>Elasmobranchia</b>	<b>Osteichthyes</b>	<b>Invertebrata</b>
Chi-Square	$p < 0.001$	NS	NS

**Table 7.2.** Summary of statistical analyses of frequency of occurrence by sub-region for 2008 elasmobranch subgroups, showing both significant and non-significant (NS) differences and associated p-values. Determination of significance is based on  $p < 0.05$ .

2008 Mean Abundance by Sub-region - Elasmobranch Subgroups		
	<b>sharks</b>	<b>guitarfish</b>
Chi-Square	$p = 0.003$	NS

**Table 7.3.** Summary of statistical analyses of frequency of occurrence by sub-region for 2008 finfish subgroups, showing both significant and non-significant (NS) differences and associated p-values. Determination of significance is based on  $p < 0.05$ .

2008 Mean Abundance by Sub-region - Finfish Subgroups					
	<b>demersal fishes</b>	<b>inshore fishes</b>	<b>midwater/pelagic fishes</b>	<b>reef fishes</b>	<b>flatfishes</b>
Chi-Square	$p = 0.038$	$p = 0.012$	NS	$p = 0.007$	$p = 0.024$

**Table 7.4.** Summary of statistical analyses of frequency of occurrence by sub-region for 2008 invertebrate subgroups, showing both significant and non-significant (NS) differences and associated p-values. Determination of significance is based on  $p < 0.05$ .

2008 Mean Abundance by Sub-region - Invertebrate Subgroups						
	<b>squids, octopus</b>	<b>bivalves, gastropods</b>	<b>crabs, shrimps</b>	<b>echinoderm</b>	<b>jellyfish</b>	<b>sessile inverts</b>
Chi-Square	NS	NS	$p = 0.015$	NS	$p = 0.031$	$p < 0.001$

**Table 7.5.** Frequency of occurrence of trawls with at least one prey group present from 2000-2003 and 2008 by region.

Year	Region	Prey Species	
		Present	Absent
2000	Northern	74.6	25.4
	Central	82.6	17.4
	Southern	78.7	21.3
	<b>Total</b>	<b>78.1</b>	<b>21.9</b>
2001	Northern	79.8	20.2
	Central	93.5	6.5
	Southern	95	5.0
	<b>Total</b>	<b>89.7</b>	<b>10.3</b>
2002	Northern	56.6	43.4
	Central	88.6	11.4
	Southern	91.3	8.7
	<b>Total</b>	<b>78.2</b>	<b>21.8</b>
2003	Northern	67.9	32.1
	Central	82	18.0
	Southern	51.9	48.1
	<b>Total</b>	<b>66.4</b>	<b>33.6</b>
2008	Northern	72.6	27.4
	Central	77.2	22.8
	Southern	76.4	23.6
	<b>Total</b>	<b>75.2</b>	<b>24.8</b>

**Table 7.6.** Classification of bottom habitat expressed as percentages by year and region.

Year	Region	Not Hard Bottom	Probable Hard Bottom	Hard Bottom
2000	Northern	80.4	15.0	4.6
	Central	82.6	12.3	5.2
	Southern	81.8	12.9	5.3
	<b>Total</b>	<b>81.5</b>	<b>13.5</b>	<b>5.0</b>
2001	Northern	87.6	6.2	6.2
	Central	94.1	4.7	1.2
	Southern	88.3	9.6	2.1
	<b>Total</b>	<b>89.7</b>	<b>7.1</b>	<b>3.2</b>
2002	Northern	93.8	5.0	1.2
	Central	95.0	4.5	0.5
	Southern	92.5	6.7	0.8
	<b>Total</b>	<b>93.7</b>	<b>5.4</b>	<b>0.9</b>
2003	Northern	88.2	7.0	4.8
	Central	96.0	3.5	0.5
	Southern	95.1	4.1	0.8
	<b>Total</b>	<b>92.7</b>	<b>5.0</b>	<b>2.2</b>
2008	Northern	90.7	6.0	3.3
	Central	78.4	17.3	4.3
	Southern	95.3	3.3	1.4
	<b>Total</b>	<b>89.0</b>	<b>8.1</b>	<b>2.9</b>

**Table 7.7.** Frequency of collection (%) of loggerheads in trawl events by sub-region.

Region	Loggerhead	
	Present	Absent
Northern	19.6	80.4
Central	19.1	80.9
Southern	29.1	70.9
<b>Overall</b>	<b>22.8</b>	<b>77.2</b>

**Table 7.8.** Summary of logistic regression analyses of loggerhead occurrence and bottom habitat and individual prey species for the northern sub-region. Determination of significance is based on  $p < 0.05$ .

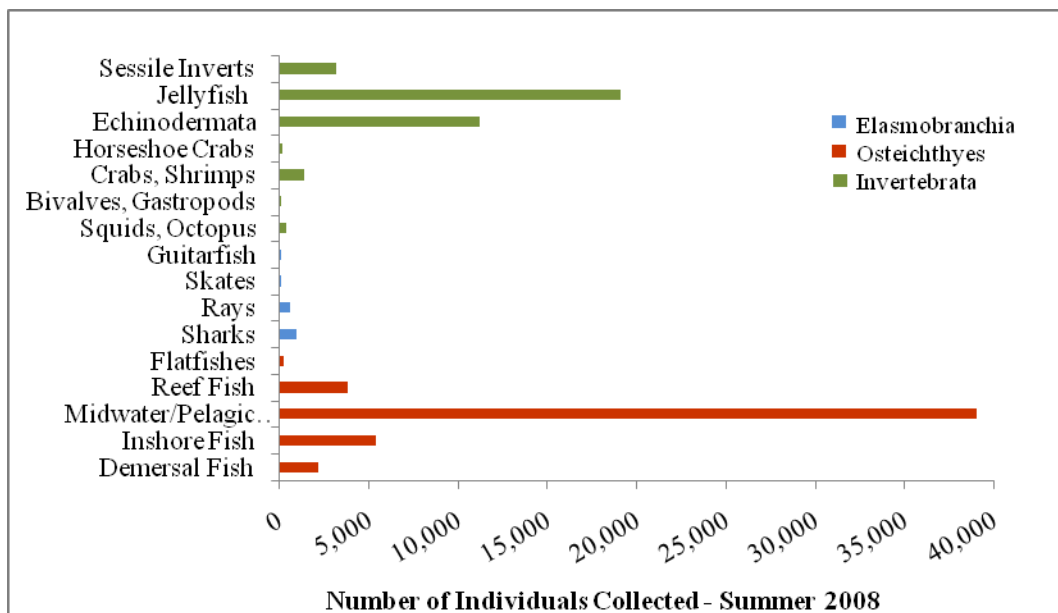
<b>Northern Sub-region</b>			
Model Chi Square = 0.057			
Attribute	Log-odds	Odds Ratio	P-value
Bottom Type	-0.091	0.913	0.599
Portunid Crab	-0.549	0.578	0.059
Spider Crab	-0.713	0.49	<b>0.002</b>
Misc Crab	-0.07	0.932	0.777
Whelk/Conch	-1.004	0.366	0.341
Misc. Jellyfish	-0.005	1.005	0.977
Blue Crab	-0.144	0.866	0.702
Horseshoe Crab	-0.17	0.843	0.64
Stone Crab	-0.028	0.972	0.933
Cannonball Jellyfish	0.285	1.33	0.09

**Table 7.9.** Summary of logistic regression analyses of loggerhead occurrence and bottom habitat and loggerhead occurrence and individual prey species for the central sub-region. Determination of significance is based on  $p < 0.05$ .

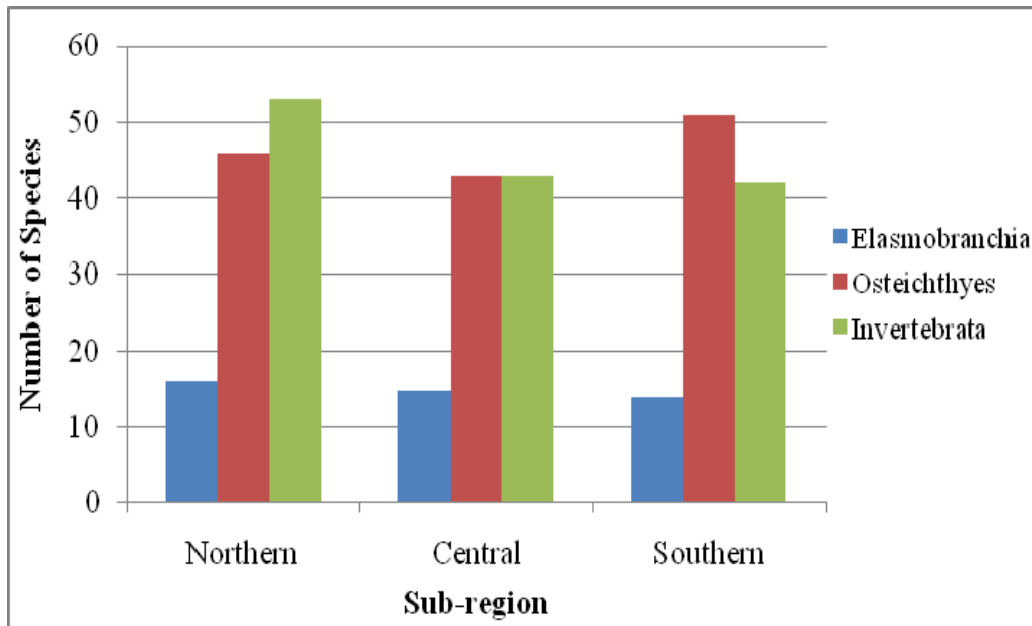
<b>Central sub-region</b>			
Model Chi Square = 0.003			
Attribute	Log-odds	Odds Ratio	P-value
Bottom Type	-0.314	0.731	0.232
Portunid Crab	-0.277	0.797	0.395
Spider Crab	-0.514	0.598	<b>0.043</b>
Misc Crab	-0.222	0.801	0.326
Whelk/Conch	0.204	1.227	0.694
Misc. Jellyfish	-0.564	0.569	<b>0.004</b>
Blue Crab	-0.165	0.848	0.651
Horseshoe Crab	-0.444	0.642	0.374
Stone Crab	-0.113	0.893	0.673
Cannonball Jellyfish	-0.201	0.818	0.28

**Table 7.10.** Summary of logistic regression analyses of loggerhead occurrence and bottom habitat and loggerhead occurrence and individual prey species for the southern sub-region. Determination of significance is based on  $p < 0.05$ .

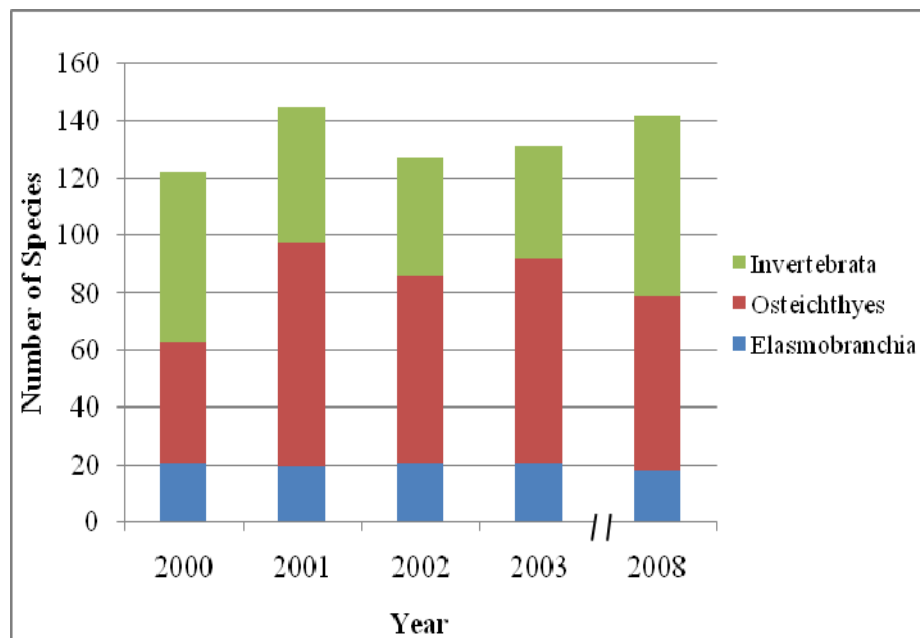
Southern sub-region			
Model Chi Square = 0.000			
Attribute	Log-odds	Odds Ratio	P-value
Bottom Type	-0.098	0.906	0.598
Portunid Crab	-0.357	0.7	0.06
Spider Crab	-0.958	0.384	<b>0.000</b>
Misc Crab	0.016	1.016	0.936
Whelk/Conch	-0.017	0.983	0.973
Misc. Jellyfish	-0.249	0.780	0.075
Blue Crab	-0.297	0.743	0.337
Horseshoe Crab	0.011	1.011	0.972
Stone Crab	-0.402	0.669	0.101
Cannonball Jellyfish	0.144	1.155	0.324



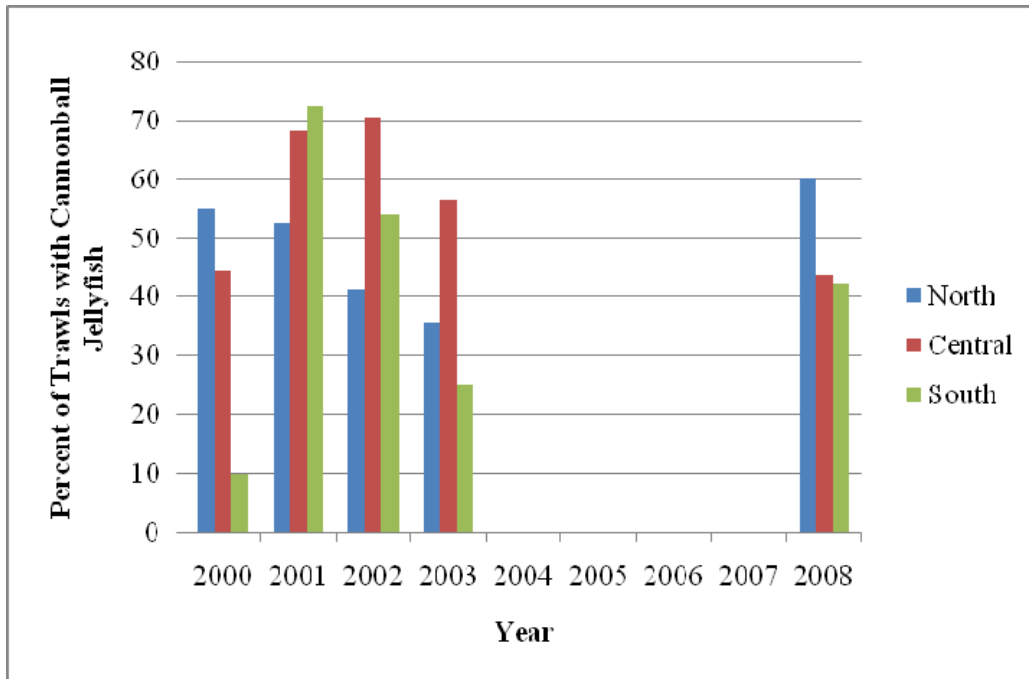
**Figure 7.1.** Total abundance (number of individuals) of by-catch subgroups collected in trawls during May-July 2008.



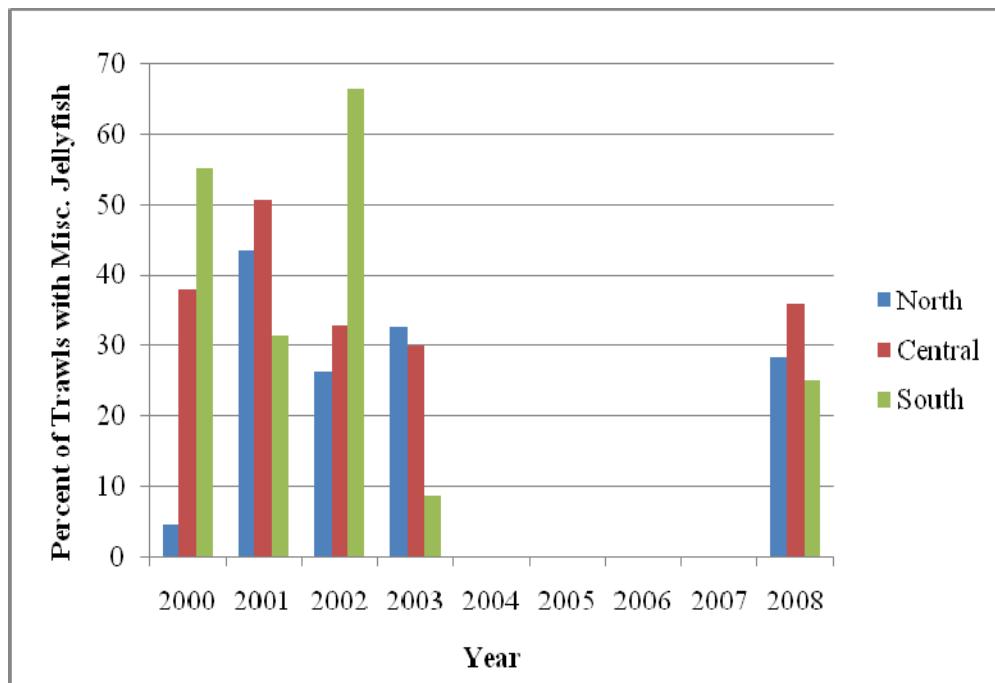
**Figure 7.2.** Species number of by-catch groups collected in trawls in the northern, central, and southern sub-regions during May-July 2008.



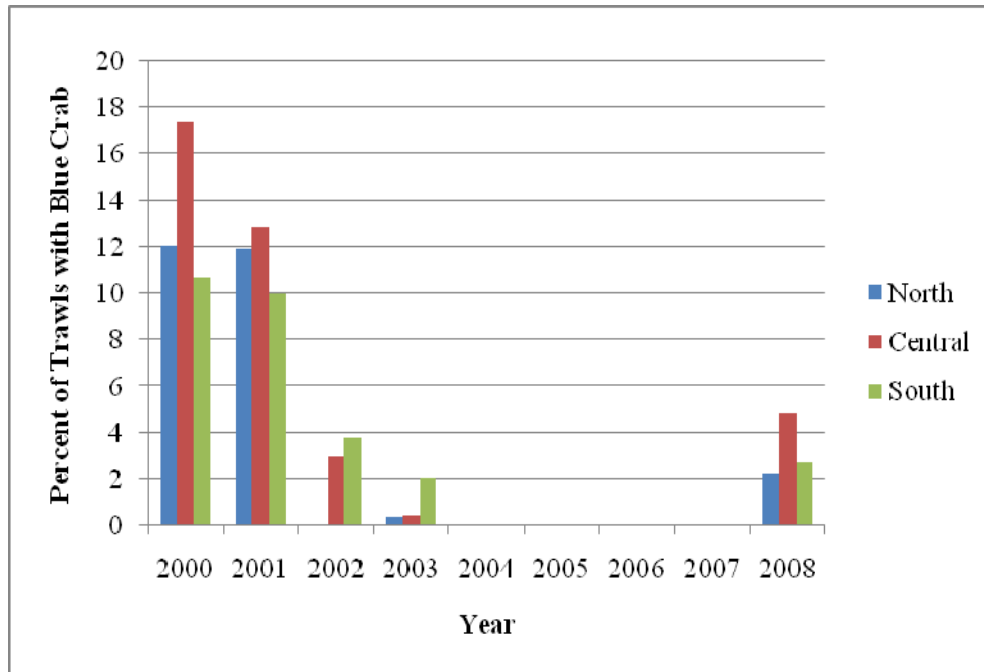
**Figure 7.3.** Total number of species collected in trawls during 2000-2003 and 2008, as well as species number for each major by-catch group.



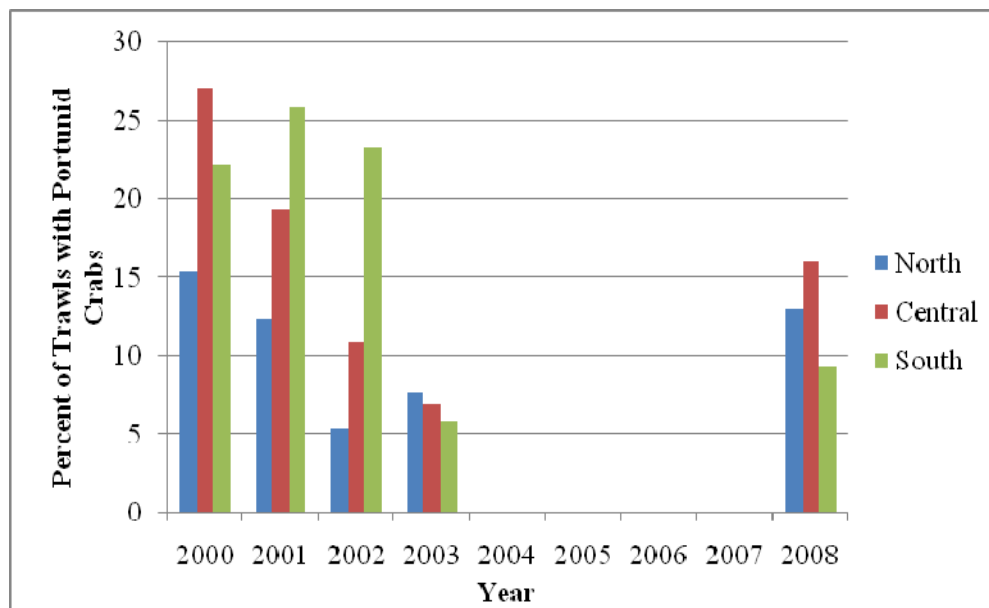
**Figure 7.4.** Frequency of occurrence of trawls with cannonball jellyfish present during 2000-2003 and 2008 for each sub-region.



**Figure 7.5.** Frequency of occurrence of trawls with miscellaneous jellyfish present during 2000-2003 and 2008 for each sub-region.

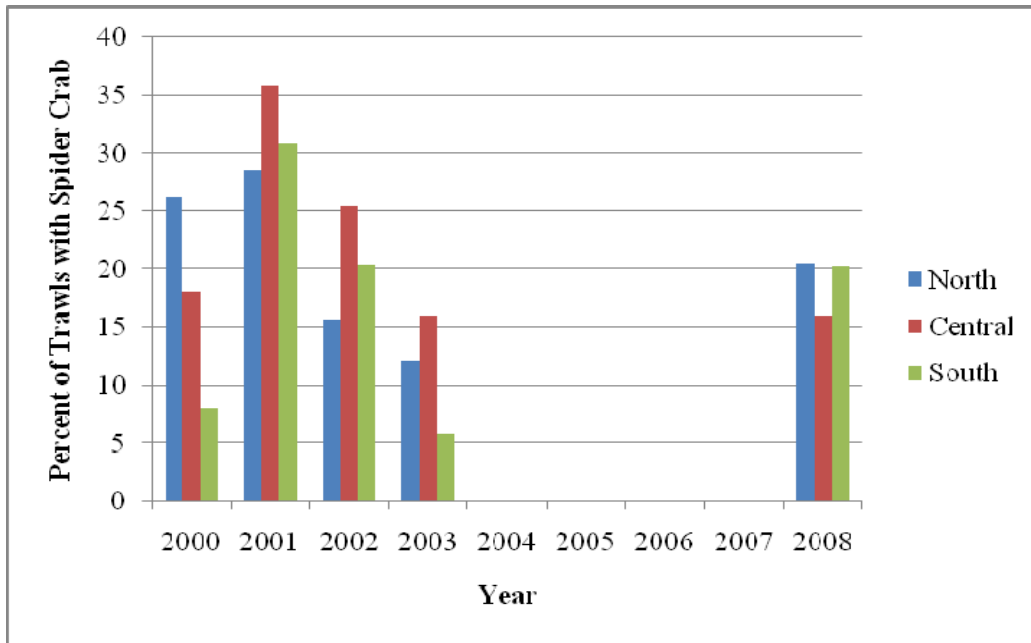


**Figure 7.6.** Frequency of occurrence of trawls with blue crab present during 2000-2003 and 2008 for each sub-region.

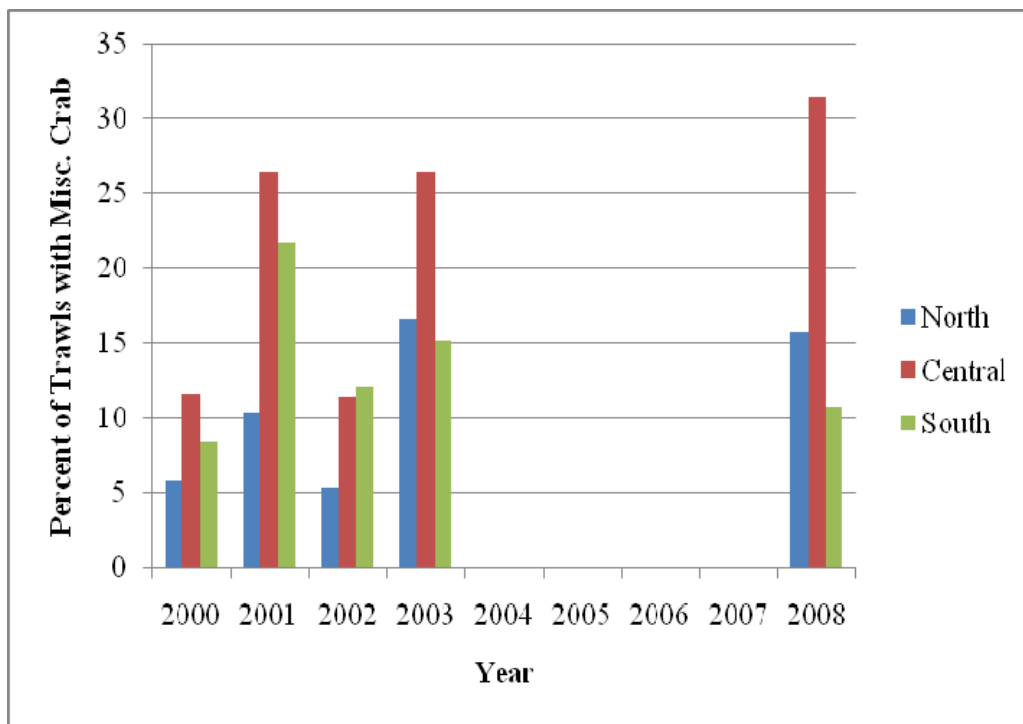


**Figure 7.7.** Frequency of occurrence of trawls with Portunid crab present during 2000-2003 and 2008 for each sub-region.

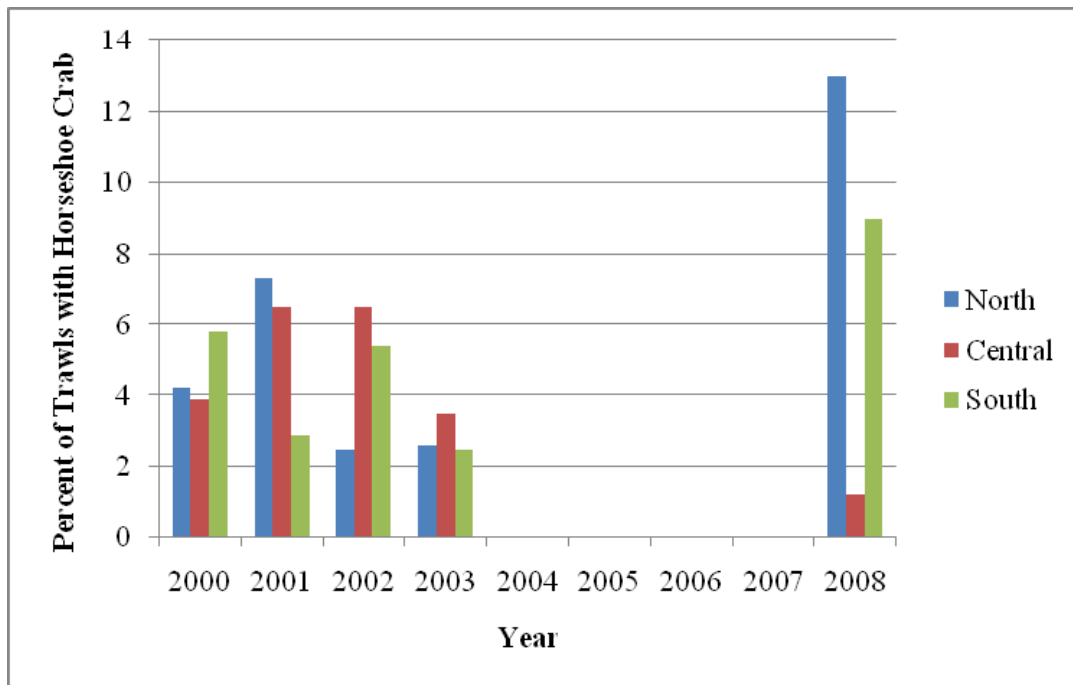




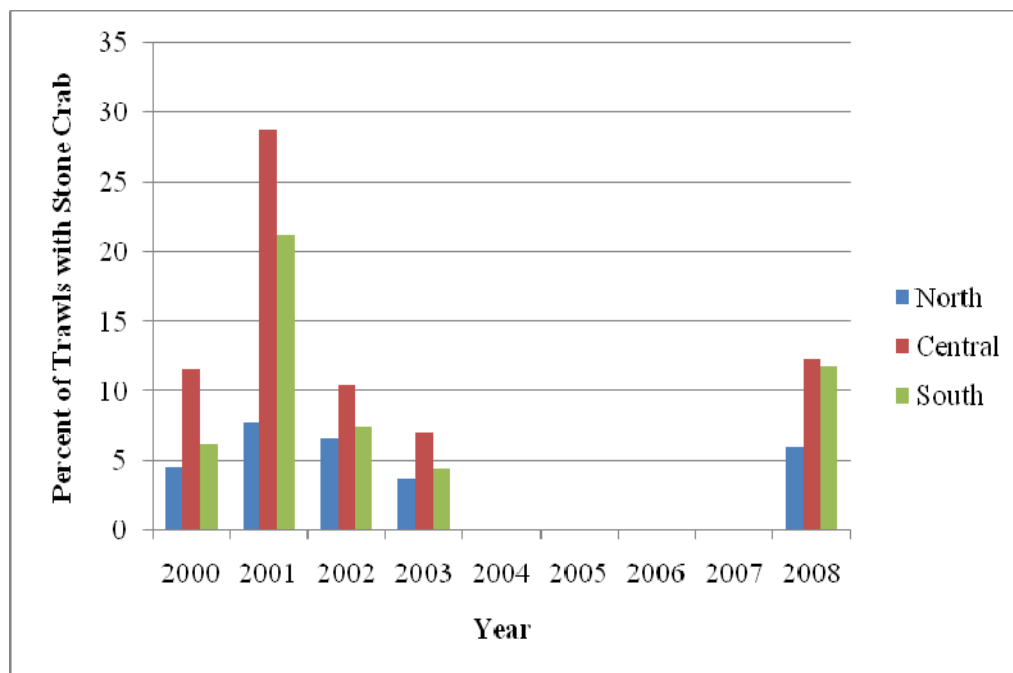
**Figure 7.8.** Frequency of occurrence of trawls with spider crab present during 2000-2003 and 2008 for each sub-region.



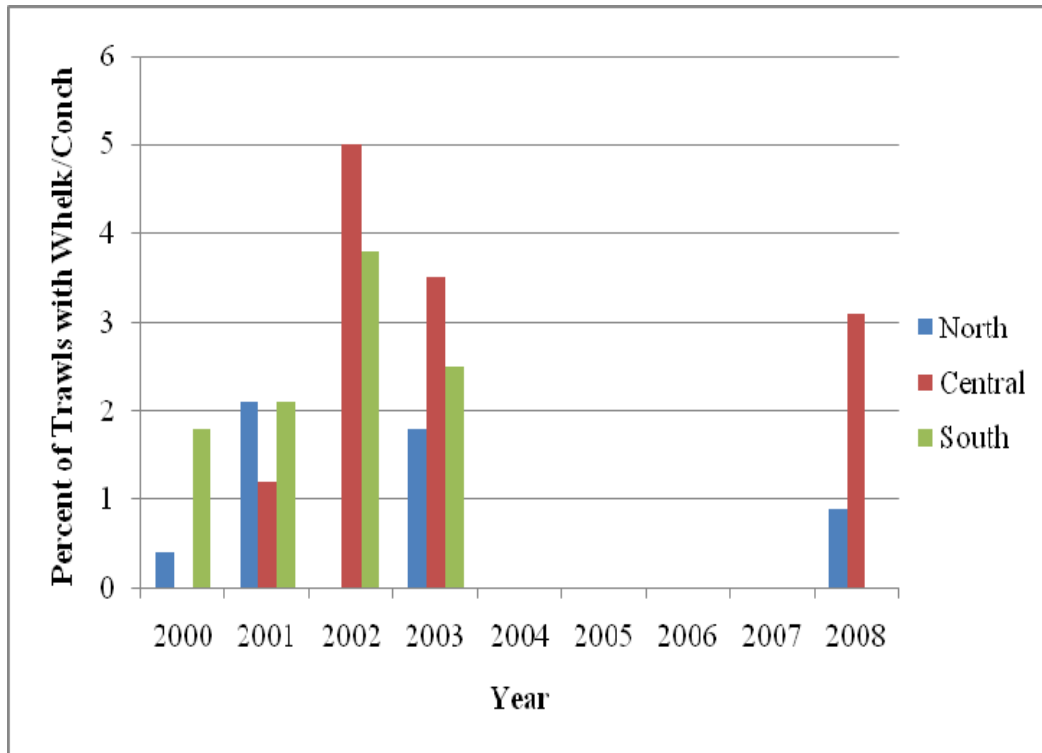
**Figure 7.9.** Frequency of occurrence of trawls with miscellaneous crab present during 2000-2003 and 2008 for each sub-region.



**Figure 7.10.** Frequency of occurrence of trawls with horseshoe crab present during 2000-2003 and 2008 for each sub-region.



**Figure 7.11.** Frequency of occurrence of trawls with stone crab present during 2000-2003 and 2008 for each sub-region.



**Figure 7.12.** Frequency of occurrence of trawls with whelk/conch present during 2000-2003 and 2008 for each sub-region.

## Chapter 8 Status Report for the 2008 Endocrine Disruption Study (EDS)

In summer 2008, a multi-collaborator effort was initiated to evaluate the degree to which the endocrine systems of juvenile loggerheads (<70cm SCLmin) may be compromised as a result of exposure to a suite of contaminants. Two spatially distinct coastal study areas (Brunswick, GA, and Charleston, SC) were selected based on their relative location to discharge from urbanized and industrially developed port cities, and subsequently their proximity to the homeports for research vessels conducting the sea turtle trawl survey (given the time-sensitive analysis requirements for refrigerated sampled).

Nearly 200 chemical compounds from eight chemical classes (Table 8.1) have been identified for consideration in this study. Contaminant analyses will be conducted by Dr. Jennifer Keller and Mr. Jared Ragland at National Institute of Standards and Technology (NIST) in Charleston, SC. A suite of parameters that collectively enable assessment of endocrine system functionality will be analyzed by numerous collaborators (Table 8.2).

In 2008, 105 samples (64 from Brunswick and 41 from Charleston) were collected for various contaminant studies. Some samples were collected from reproductively mature loggerheads and therefore will not be utilized for the EDS. Similarly, not all samples collected for this collaborator and/or other collaborators were determined to be viable, further reducing the number of samples which can be used. However, it is highly probable that the goal of 25 viable samples from each study location will be analyzed.

Methods for analyzing contaminant samples are still under development; however, finalization of methods is anticipated in summer 2009 with data available in spring 2010.

To date, samples have been provided to all collaborators except for thyroid samples which still need to be shipped to Dr. Duncan MacKenzie (Texas A&M University). Samples have been analyzed and results made available for testosterone, corticosterone, estradiol, selected vitamins, genetics, mercury, and blood chemistry. Sex ratio (testosterone) and genetics data are presented in Chapter 1 of this report, and mercury data are discussed at length in Chapter 6. Raw data has also been provided for estradiol. Summaries of findings for corticosterone and selected vitamins are provided below.

### *Corticosterone*

Corticosterone concentrations and time before bleeding were log-transformed to meet assumptions for normality and equality of variances. Tests were run using R, version 2.6.2.

Corticosterone levels ranged 1.01-8.87 ng/mL (average  $4.68 \pm 2.3$  ng/mL,  $n=22$ ), and 1.08-15.04 ng/mL (average  $5.20 \pm 3.91$  ng/mL,  $n=39$ ) for the Charleston and Brunswick areas, respectively. Corticosterone concentrations were not statistically significant between the two areas (Welch two-sample t-test,  $t=0.6534$ ,  $df=58.859$ ,  $p=0.516$ ). To account for potential increase in plasma corticosterone with increasing time on deck before blood was collected, we looked at the relationship between these two variables. Time before bleeding on deck averaged 7.54 min (range: 2 to 25 min) for turtles collected near Charleston, SC, and 7.66 min (range: 2 to 39 min) for turtles collected near Brunswick, GA. Time before bleeding for turtles collected in the

Charleston area was not significantly different from that of turtles collected in the Brunswick area (Welch two-sample t-test,  $t=-0.09$ ,  $df=42$ ,  $p=0.92$ ). Moreover, a significant relationship between the time elapsed before bleeding and corticosterone levels was not detected (Kendall's rank correlation,  $\tau = 0.0046$ ,  $p = 0.96$ ).

Blood samples for 77% ( $n=47$ ) of all turtles collected in this study were taken within 10 minutes, and variability in corticosterone levels was high within these 10 minutes (mean=4.79 ng/mL, range: 1.01-15.04 ng/mL). This reflects the fact that corticosterone levels can greatly vary depending on a number of factors, including: health and/or physical status (Snoddy et al., in press), time trapped underwater in the net (Snoddy et al., in press) and reproductive status and season (Gregory et al., 1996; Valverde et al., 1999; Jessop, 2001; Blanvillain et al., 2008).

The fact that we did not find a significant relationship between time before bleeding and corticosterone might reflect the relatively short time window during which blood samples were collected. Indeed, Gregory et al. (1996) noted an increase in corticosterone levels after 1h, and then levels reached a peak at 3h and started decreasing after 6h. Moreover, corticosterone levels measured by Gregory et al. (1996) on loggerheads <80cm SCLmin collected by trawls had mean concentrations of 4.71ng/mL which is comparable to what we measured in this study. Finally, levels reported in our study appear relatively low when compared to those of juvenile green turtles captured in gillnets, with corticosterone ranging from 0.3-52ng/mL (average: 20.8 ng/mL), possibly showing species-specific difference in corticosterone response to stress of capture. Lower corticosterone levels measured in our study when compared to those of Snoddy et al. (in press) show a difference in stress response based on the capture technique.

### ***Vitamins A & E***

The Georgia Sea Turtle Center is leading an effort to develop a baseline data base for loggerhead sea turtle nutritional levels. We are also exploring the possibility of using plasma vitamin A as a biomarker for organochlorine pesticide exposure. Organochlorines inhibit vitamin A synthesis. Samples have been taken from turtles captured on the RV *Georgia Bulldog*, nesting female loggerheads on Jekyll Island, and ill and injured loggerheads presenting to the GSTC for rehabilitation. The turtles in rehabilitation are being monitored sequentially throughout their recovery period. To date, we have only measured levels of plasma vitamin A and E. The remainder of the samples will be processed in the near future. A preliminary data summary (Table 8.3) indicates that:

- Retinol (Vitamin A) concentrations were significantly higher in rehabilitated animals than in wild animals.
- Considering rehabilitation animals alone, retinol was significantly lower at entry and showed a positive correlation with time in rehabilitation.
- There were no significant differences or correlations associated with alpha tocopherol (Vitamin E) concentrations.

**Table 8.1.** Contaminants and contaminant classes to be analyzed for the EDS study.

Chemical Class	N compounds
Polychlorinated biphenyls (PCBs)	86
OH-PCBs	28
Polybrominated diphenyl ethers (PBDEs)	28
OH-PDBEs	13
Hexabromocyclododecanes (HBCD)	3
DDTs	6
Toxaphene	23
Other Organochlorine Pesticides (OCPs)	12

**Table 8.2.** Collaborators coordinating the analysis of endocrine indicators, additional contaminants and demographic data for a holistic study of foraging, nutrition and health.

Endocrine Indicator	Collaborator	Affiliation	Location
Testosterone	Owens/Blanvillain	College of Charleston	Charleston, SC
Coritcosterone	Owens/Blanvillain	College of Charleston	Charleston, SC
Estrogen	Rostal	Georgia Southern University	Statesboro, GA
Vitellogenin	Valverde	Southeastern Louisiana University	Hammond, LA
Thyroid Hormone	MacKenzie	Texas A&M University	College Station, TX
Retinol / Vitamins	Norton	Georgia Sea Turtle Center	Jekyll Island, GA
Blood Chemistry	Antech Diagnostics	data housed by SCDNR	Charleston, SC
Lysozyme	Peden-Adams	Medical University of South Carolina	Charleston, SC
Other variables	Collaborator	Affiliation	Location
Blood/Scute Hg	Day	National Institute of Standards and Technology	Charleston, SC
Stable Isotopes	Reich	University of Florida	Gainesville, FL
Genetics	Quattro	University of South Carolina	Columbia, SC

**Table 8.3.** Descriptive statistics for preliminary analyses of Vitamins A & E.

Status	N	Mean	Std	Min	Max
Rehab Vitamin A	28	0.65	0.23	0.2	0.98
Rehab Vitamin E	28	7.24	5.34	ND	31.8
Wild Vitamin A	28	0.51	0.14	0.24	0.87
Wild Vitamin E	28	6.32	2.39	3.09	11.28

Status	N	Mean	Std	Min	Max
Entry Vitamin A	5	0.35	0.21	0.2	0.74
Entry Vitamin E	5	3.48	2.13	ND	5.49
Rehab Vitamin A	23	0.72	0.19	0.28	0.98
Rehab Vitamin E	23	8.06	5.5	4.29	31.8

## **Chapter 9 Veterinary Assistance and Loggerhead Rehabilitation**

Ethical and humane treatment of animal subjects has become a mainstay of many research studies throughout the United States and aboard; however, the importance of this dimension of science is especially significant for projects working with endangered species like sea turtles.

Since 2000, veterinary assistance has been fully integrated into the research model utilized by this study. Veterinarian-approved data collection methods were either adopted or incorporated into the study with direct consultation of veterinarians. Rising second and third year veterinary students from the College of Veterinary Medicine at North Carolina State University (NCSU) participated in every year of the regional trawl survey (2000-2003, 2008) and in the first two years of the Charleston channel survey. In 2007, a resident radiologist (Dr. Anthony Pease, D.V.M.) greatly enhanced reproductive data collection efforts with adult male loggerheads in Port Canaveral, FL. Lastly, a most opportune collaboration has burgeoned in recent years between the in-water sea turtle trawl survey and two facilities (South Carolina Aquarium, Georgia Sea Turtle Center) that promote sea turtle rehabilitation and are located in the research vessel homeports. Between 2004 and 2008, seven (Table 9.1) of 575 loggerheads collected were determined to be in need of shore-based care and were successfully treated at these two facilities.

Five juvenile loggerheads collected from the Charleston, SC, shipping entrance channel between 2004 and 2007 were transferred to the South Carolina Aquarium (SCA). The first loggerhead transferred to the SCA was the victim of interaction with a boat propeller and had exposed lung tissue (Figure 9.1); however, wounds were successfully treated and this turtle was released 11 months later. Two juveniles were treated for “Debilitated Turtle Syndrome (DBS)”, which was caught in the early stage of onset and enabled these turtles to be released after only 3-4 months in rehabilitation (Figure 9.2). Both were subsequently tracked via satellite-telemetry (one by SCDNR, the other by the SCA), and data generally indicated typical distribution and behavior post-release. Two juveniles were treated for stingray spine wounds which occurred during their collection in the trawling gear. In both instances, training by a veterinarian (Dr. Al Segars, D.V.M.) enabled personnel to successfully remove the entire stingray barb from the wound while inflicting minimal damage to the turtle. One stingray wound was minor, enabling turtle release within a month of rehabilitation, and this turtle was also monitored post-release via satellite telemetry. The second stingray wound was more severe, with necrotic fascitis resulting from an allergic reaction to the stingray barb venom (Figure 9.3); this turtle was held through winter 2008 and will be released in spring 2009. Stingray wound treatment procedures have subsequently been modified to include IM injection of Dexamethasone (0.5 mg/kg) upon barb removal, per the recommendation of the Head Veterinarian at the SCA, Dr. Shane Boylan, D.V.M.

Two juvenile loggerheads collected in summer 2008 in northern FL were transferred to the Georgia Sea Turtle Center (GSTC) on Jekyll Island, GA, for treatment of boat strike wounds. Wounds (broken and dead bone near the peak of the carapace) were less severe for one juvenile (“Gale”) admitted to the GSTC, enabling release after 3.5 months. The GSTC attached a satellite transmitter (using extra epoxy to potentially increase track duration) to Gale before being released on 4 October 2008. Gale over-wintered south of Jacksonville, FL, in close proximity to where collected, and continues to be detected daily. The second juvenile (“Duffy”) had a large slice through the shell (carapace, bridge and plastron) and an even larger wound on the right rear

flipper revealing a broken femur. Deep wounds were treated with honey, and bees wax was packed into the wounds to protect them. Honey has been used in human medicine for centuries and is now being used for wounds that are difficult to heal such as those found in diabetic patients. Furthermore, the GSTC is working with the local bee keeping community to obtain the bees wax and honey. Wound healing was dramatic after administering honey as a treatment, and “Duffy” is anticipated to be released in summer 2009. Personnel from SCDNR also assisted in satellite tagging a third juvenile loggerhead (“Golden Boy”) at the GSTC (their first) in July 2007, although this loggerhead was not collected by our study.

In addition to seven loggerheads which received rehabilitation, four juvenile loggerheads were collected dead between 2004 and 2008. One dead juvenile was collected from the Port Canaveral, FL, shipping entrance channel in 2006 (Figure 9.4) and two others were collected from the same location in 2007. All three loggerheads collected from the Port Canaveral channel had been dead for awhile given a distinct odor. In contrast, a freshly killed juvenile loggerhead was collected from the Charleston, SC, shipping entrance channel in May 2006 (Figure 9.5). In all four instances, loggerhead mortalities resulted from interactions with large boat props, given the complete bi-section (longitudinal as well as transverse cuts) of the turtles which resulted. The frequency of occurrence of lethal boat strike interactions is not known; however, mitigation of boat strikes is a research need and recovery objective (NMFS & USFWS, 2008).

Similar to infrequent collection of dead loggerheads, very few loggerheads tagged and released by our research activities have later been reported as stranded. Of 1,461 loggerheads collected since 2000, only seven stranding reports have been received, five of which occurred between 2004 and 2008. A nearly mature (77.8cm SCLmin) female loggerhead collected off SC in 2000 stranded on a beach near Destin, FL, in 2004. A nearly mature (80.2cm SCLmin) male loggerhead turtle collected from the Charleston, SC, shipping entrance channel in May 2004 stranded on a nearby beach the following May. One juvenile loggerhead collected in northern FL and two juvenile loggerheads collected from southern GA during 2002 and 2003 were reported stranded near Fernandina, FL, (2006) and on Cumberland Island, GA, (2006, 2008), respectively. Cause of mortality and stranding was not provided for any of these loggerheads.

Lastly, a novel deformity was noted for a juvenile loggerhead collected off FL in August 2008 (Figure 9.6). Most of the anterior upper jaw was missing, and what remained was incredibly mangled. Tactile responsiveness tests indicated full mobility of the lower jaw; however, due to the absence of an upper palate, it was unclear to what extent this turtle could grip, crush and consume crustacean prey (Lutcavage and Musick, 1985). Blood parameters measured at sea indicated normal values (Glucose = 69; Total Protein = 3.2; Hematocrit = 37); thus, after consultation with both Dr. Terry Norton, D.V.M. of the GSTC and Mrs. Megan Koperski of the FL Fish and Wildlife Commission, this turtle was released back into the wild without further treatment. Wounds such as this have been infrequently observed; however, other non-lethal wounds including bites, cracks and amputations are more commonly seen. Characterization of non-lethal wounds among sea turtles collected by our studies since 2000 is the subject of a Master’s thesis currently underway at the College of Charleston, Marine Biology Program.



**Table 9.1.** Summary of loggerheads collaboratively treated and/or monitored between the South Carolina Department of Natural Resources, University of Georgia Marine Extension Service, South Carolina Aquarium, and the Georgia Sea Turtle Center (2004-2008).

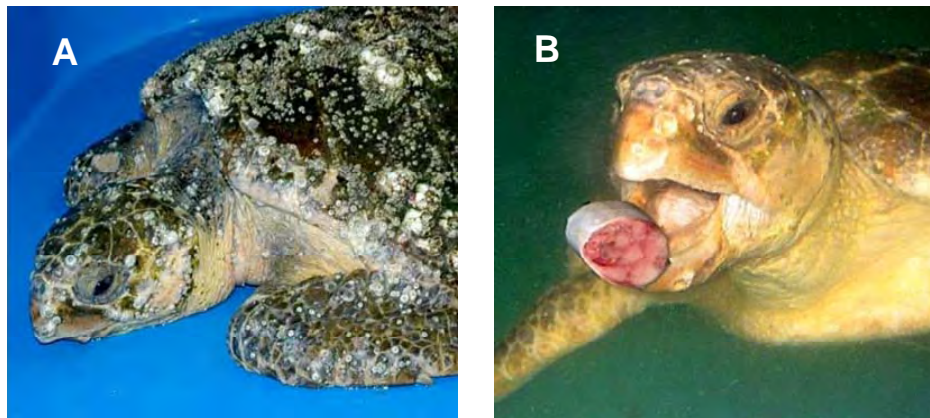
<b>Turtle ID</b>	<b>Name</b>	<b>Facility</b>	<b>Diagnosis</b>	<b>Admitted</b>	<b>Released</b>
CC0306	Channel	SCA	boat strike	6/16/2004	5/26/2005
CC0365	Jetty	SCA	DBS	5/19/2005	8/19/2005
CC0440	Lady Lisa	SCA	DBS	7/31/2007	11/3/2007
CC0408*	Stingray	SCA	puncture	8/1/2007	8/28/2007
CC0485	Little Pritchard	SCA	puncture	8/1/2008	
CC2532	Duffy	GSTC	boat strike	6/11/2005	
CC2557	Gale	GSTC	boat strike	6/25/2008	10/4/2008
n/a	Golden Boy	GSTC	DBS		7/19/2007

*\*first collected in Charleston, SC, shipping entrance channel in May 2006*

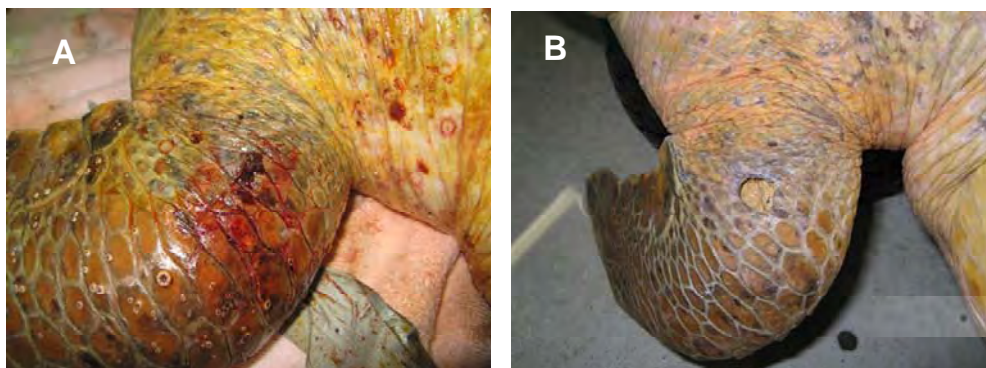
<b>Turtle ID</b>	<b>Satellite Telemetry Tracking Map</b>
CC0306	
CC0365	<a href="http://www.seaturtle.org/tracking/index.shtml?tag_id=58944">http://www.seaturtle.org/tracking/index.shtml?tag_id=58944</a>
CC0440	<a href="http://www.seaturtle.org/tracking/index.shtml?tag_id=49619">http://www.seaturtle.org/tracking/index.shtml?tag_id=49619</a>
CC0408*	<a href="http://www.seaturtle.org/tracking/index.shtml?tag_id=73120">http://www.seaturtle.org/tracking/index.shtml?tag_id=73120</a>
CC0485	
CC2532	
CC2557	<a href="http://www.seaturtle.org/tracking/index.shtml?tag_id=84652">http://www.seaturtle.org/tracking/index.shtml?tag_id=84652</a>
n/a	<a href="http://www.seaturtle.org/tracking/index.shtml?tag_id=49618">http://www.seaturtle.org/tracking/index.shtml?tag_id=49618</a>



**Figure 9.1.** Boat-strike interaction and exposed lung tissue, CC0306. Photos courtesy of Mrs. Kelly Thorvalson, SCA.



**Figure 9.2.** Before (A) and after (B) images for CC0365 rehabilitated at the SCA. Photos courtesy of Mrs. Barbara Bergwerf.



**Figure 9.3.** Before (A) and after (B) views of stingray barb puncture wound for CC0485. Photos courtesy of Dr. Shane Boylan, D.V.M. of the SCA.



**Figure 9.4.** Dorsal and ventral views of longitudinal bi-section of a juvenile loggerhead killed in the Port Canaveral, FL, shipping entrance channel in April 2006.



**Figure 9.5.** Dorsal view of transverse bi-section of a juvenile loggerhead killed in the Charleston, SC, shipping entrance channel in May 2006.



**Figure 9.6.** Novel wound/deformity observed in juvenile loggerhead (CC2643) collected off northern Florida in August 2008.

## Chapter 10 Outreach and Education

Since project inception, project staff have embraced the philosophy that “if it didn’t get reported, it didn’t happen.” Subsequently, considerable emphasis is placed on disseminating the results of data collected by this long-term research project to a variety of audiences.

Data collected by this study were presented by us or collaborating researchers at every International Sea Turtle Symposium (ISTS) held between 2005 and 2009. Two oral presentations (regional trawl survey, juvenile satellite telemetry) were delivered at the 25<sup>th</sup> ISTS in Savannah, GA. A graduate student supported by this grant (and who utilized genetics data analyzed by a sub-contractual agreement with the USC Biology Department) received the “Best Student Poster” Award at the 26<sup>th</sup> ISTS in Crete, Greece. Three oral presentations (size-frequency distribution, juvenile and adult male satellite telemetry and by-catch analysis) were presented at the 27<sup>th</sup> ISTS in Myrtle Beach, SC. A graduate student utilized contaminant data collected for a collaborating researcher in a poster presented at the 28<sup>th</sup> ISTS in Loreto, Baja California Sur, Mexico. And two graduate student posters (injury characterization, contaminant studies) utilizing data collected by this study were presented at the 29<sup>th</sup> ISTS in Brisbane, Australia. Due to SCDNR travel restrictions, only domestic travel was supported by this grant.

Although peer-reviewed publication of data are still pending, annual reports have been rich with information that has been widely distributed. Furthermore, this Final Report was developed with chapters written in manuscript format, to facilitate submission for publication later this summer.

Print media also provided regular coverage of research activities conducted by this grant. In May 2006, a two-page article on trawling and satellite telemetry studies in the Charleston, SC, shipping entrance channel was featured in the “Local/State” section of the Charleston-based *Post and Courier*. In April 2007, reproductive and satellite telemetry studies with adult male loggerheads were featured on the Front Page of *Florida Today*, the local newspaper for the Space Coast region. And in August 2008, the regional in-water survey was featured in the *Brunswick News*, as a follow-up story to an article on historic loggerhead nesting declines in GA that had appeared in the *Brunswick News* two days earlier.

Finally, project personnel deliver numerous presentations to the general public each year. Although many of these presentations are seasonally distributed, they average approximately one presentation per month annually. High visibility public presentation formats during 2004-2008 included large community events such as the SCDNR Marine Resources Division’s Open House and the Marine Division’s “Coastal Exploration Series”, with events for the latter held at both the Marine Division’s facility on Fort Johnson as well as at the South Carolina Aquarium. Project personnel also make a concerted effort to reach out in the classroom, and have delivered appropriate level presentations to students ranging from 3<sup>rd</sup> graders to high school seniors. Community-based education has also occurred via field trip format, in which the audience ranged from elementary-aged students to senior citizens. Between these various events, it is likely that we directly engaged at least 1,000 members of the general public in conversations about sea turtles and our studies over the past five years.

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It goes without saying that a study of this magnitude would not have been possible without the steadfast assistance and dedication to a universal goal of many people.

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Some of the turtle crew posing with CC2509 (ID#73106, “Cougar”) prior to release. Cougar, the largest (by weight, 380 lbs or 173 kg) of 1,461 loggerheads that we have collected since 2000, was named after the mascot of a local elementary school that “adopted” him via Seaturtle.org and subsequently developed several lesson plans about sea turtles and marine life.

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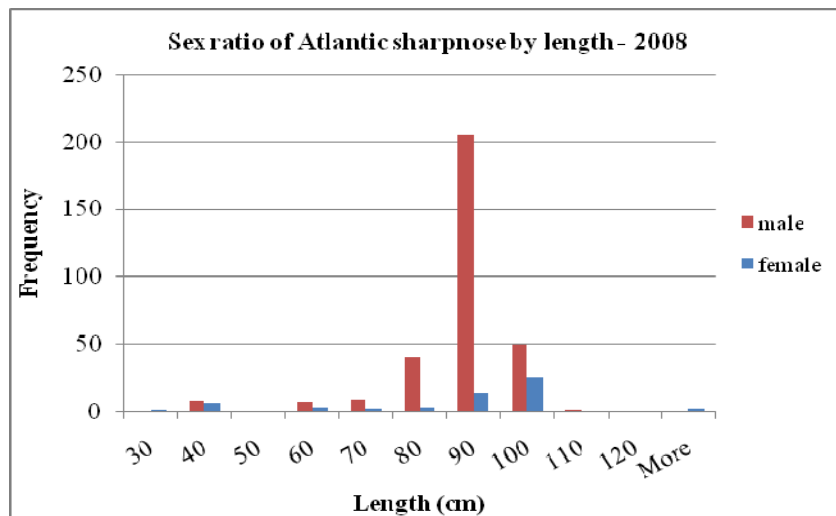
## APPENDIX A - Elasmobranchia

**Table A1.** Abundance, size ranges, and sex ratios of elasmobranch by-catch collected in trawls during 2008. Minimum, maximum, and mean sizes are in centimeters. Sex ratios are reported only for species with data recorded for  $\geq 25$  individuals.

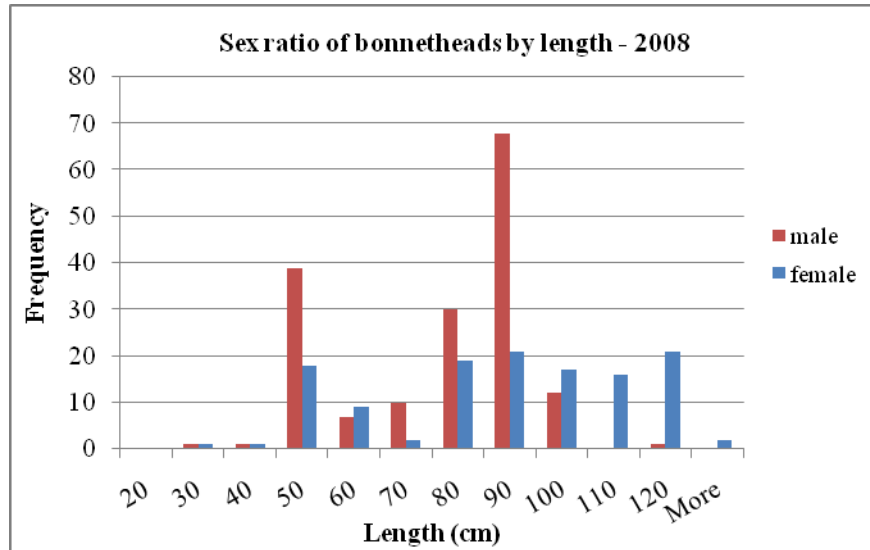
Sp Code	Sharks	Common Name	# Indiv.	Min Size	Max Size	Mean Size	Sex Ratio (M : F : U)
A005	<i>Carcharias taurus</i>	sand tiger shark	3				
A014	<i>Carcharhinus acronotus</i>	blacknose shark	68	100	150	121.00	1 : 5 : 3.7
A018	<i>Carcharhinus limbatus</i>	blacktip shark	11	80	100	93.00	
A023	<i>Galeocerdo cuvieri</i>	tiger shark	1				
A025	<i>Mustelus canis</i>	smooth dogfish	1	67	67	67.00	
A028	<i>Rhizoprionodon terraenovae</i>	Atlantic sharpnose	437	30	140	83.09	5.9 : 1 : 1
A029	<i>Sphyrna lewini</i>	scalloped hammerhead	25	42	117	55.09	5.5 : 2 : 1
A031	<i>Sphyrna tiburo</i>	bonnethead	411	23	881	80.08	1.7 : 1.3 : 1
	<b>Rays</b>						
A048	<i>Dasyatis americana</i>	southern stingray	305	14	99	41.28	1.7 : 1.5 : 1
A049	<i>Dasyatis centroura</i>	rougtail stingray	28	51	75	63.20	2 : 1 : 6.3
A051	<i>Dasyatis sayi</i>	bluntnose stingray	22	17	64	28.06	
A054	<i>Gymnura micrura</i>	smooth butterfly ray	169	24	86.3	44.48	5.4 : 7.7 : 1
A056	<i>Aetobatus narinari</i>	spotted eagle ray	10				
A057	<i>Myliobatis freminvillei</i>	bullnose ray	36	30	108	39.21	2.4 : 3.8 : 1
A059	<i>Rhinoptera bonasus</i>	cownose ray	23	35	91	51.88	
A644	<i>Mobula hypostoma</i>	devil ray	4	59	89	72.25	
	<b>Skates</b>						
A043	<i>Raja eglanteria</i>	clearnose skate	28	23	42	33.16	1 : 6.5 : 6.5
	<b>Guitarfish</b>						
A039	<i>Rhinobatos lentiginosus</i>	Atlantic guitarfish	12	49	75	57.30	

**Table A2.** Summary of statistical analyses of CPUE for the elasmobranch group and the 3 most abundant elasmobranch species among study years, showing both significant and non-significant (NS) differences and associated p-values. Determination of significance is based on  $p < 0.05$ .

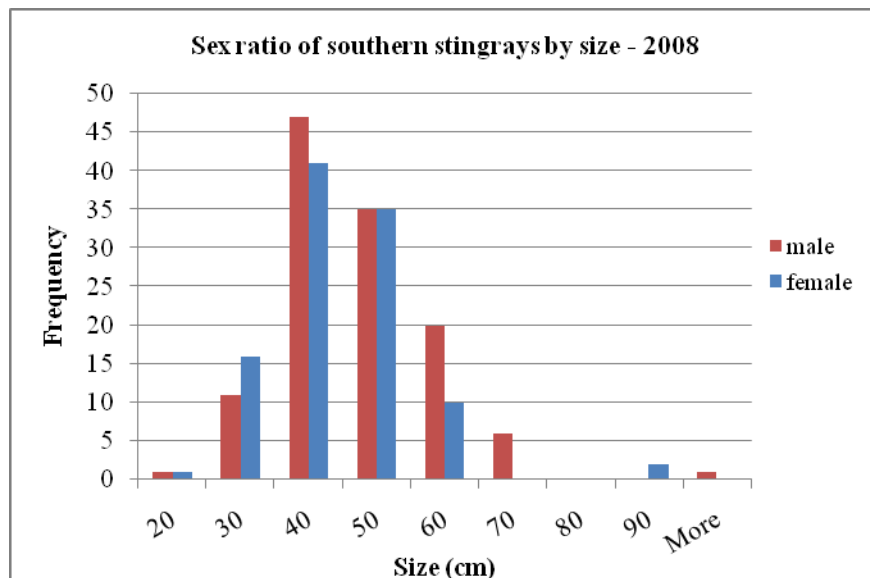
CPUE by Year				
	Elasmobranchia (overall)	Atlantic sharpnose	bonnethead	southern stingray
Kruskal-Wallis	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$
Mann-Whitney	2001>2000; $p < 0.001$	NS	2001>2000; $p < 0.001$	NS
	NS	NS	NS	2002>2000; $p < 0.001$
	2003>2000; $p < 0.001$	2003>2000; $p < 0.001$	2003>2000; $p < 0.001$	2003>2000; $p < 0.001$
	2008>2000; $p < 0.001$	2008>2000; $p < 0.001$	2008>2000; $p < 0.001$	2008>2000; $p < 0.001$
	2001>2002; $p < 0.001$	NS	2001>2002; $p = 0.002$	2002>2001; $p < 0.001$
	NS	2003>2001; $p < 0.001$	NS	2003>2001; $p < 0.001$
	2008>2001; $p < 0.001$	2008>2001; $p < 0.001$	NS	2008>2001; $p < 0.001$
	2003>2002; $p < 0.001$	2003>2002; $p < 0.001$	2003>2002; $p = 0.024$	2003>2002; $p < 0.001$
	2008>2002; $p < 0.001$	2008>2002; $p < 0.001$	2008>2002; $p = 0.010$	2008>2002; $p = 0.017$
	2008>2003; $p = 0.004$	2008>2003; $p < 0.001$	NS	NS



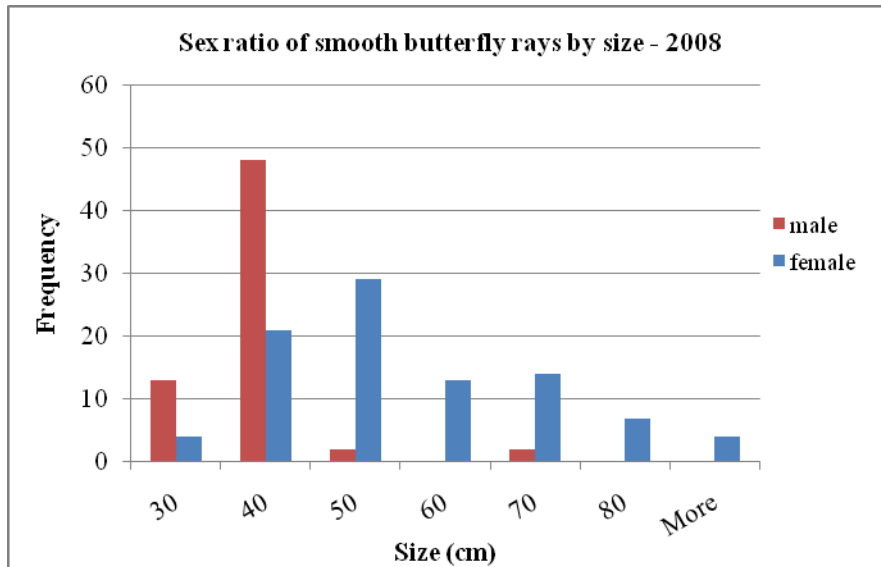
**Figure A1.** Sex ratio by length of Atlantic sharpnose sharks (*Rhizoprionodon terraenovae*) caught in trawls during 2008. Lengths are in centimeters.



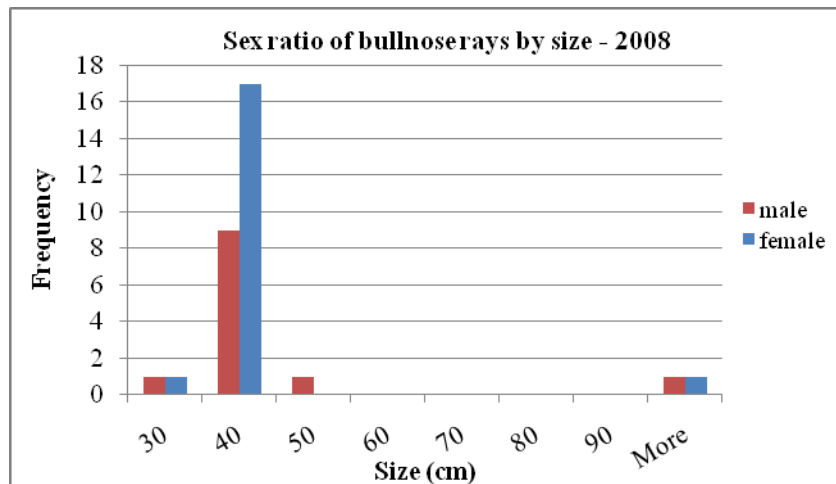
**Figure A2.** Sex ratio by length of bonnethead sharks (*Sphyrna tiburo*) caught in trawls during 2008. Lengths are in centimeters.



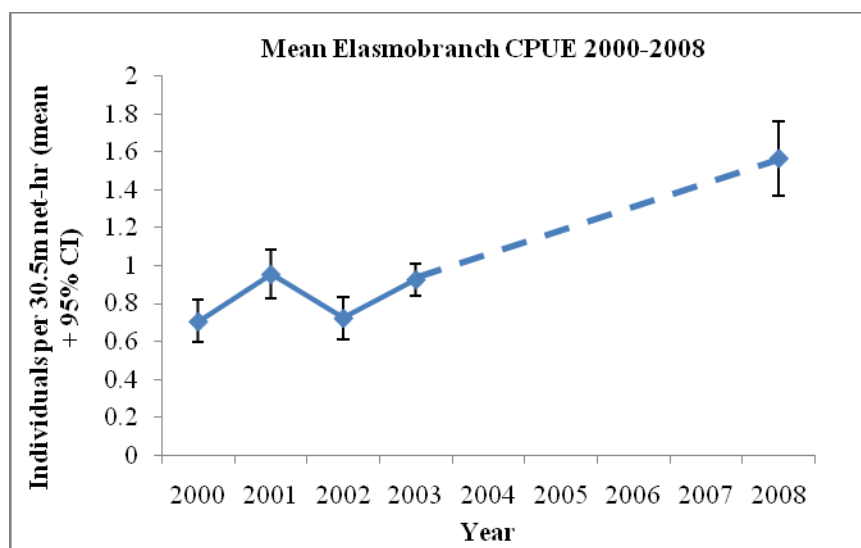
**Figure A3.** Sex ratio by size of southern stingrays (*Dasyatis americana*) caught in trawls during 2008. Sizes are in centimeters.



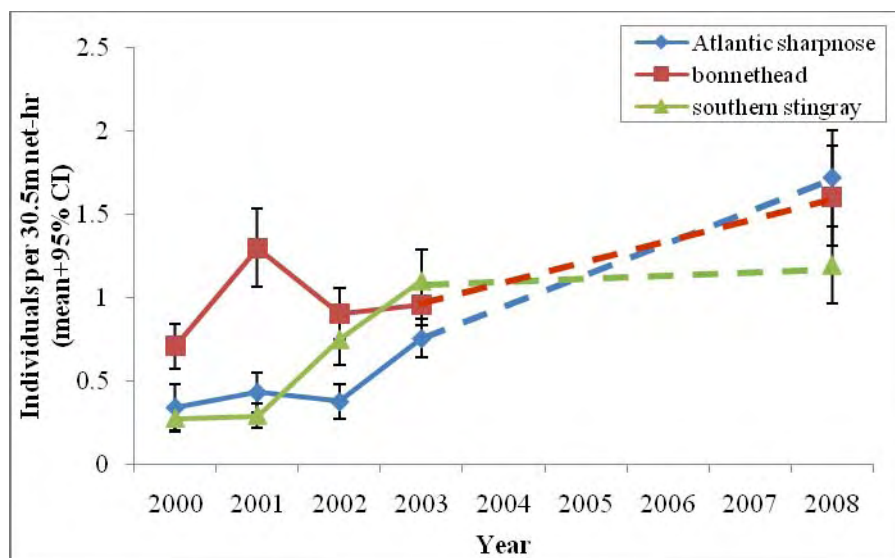
**Figure A4.** Sex ratio by size of smooth butterfly rays (*Gymnura micrura*) caught in trawls during 2008. Sizes are in centimeters.



**Figure A5.** Sex ratio by size of bullnose rays (*Myliobatis freminvillei*) caught in trawls during 2008. Sizes are in centimeters.



**Figure A6.** Mean CPUE for elasmobranchs collected in trawls in 2000-2003 and 2008. Error bars represent 95% confidence intervals. See Table 7.A2 for results of statistical analyses.

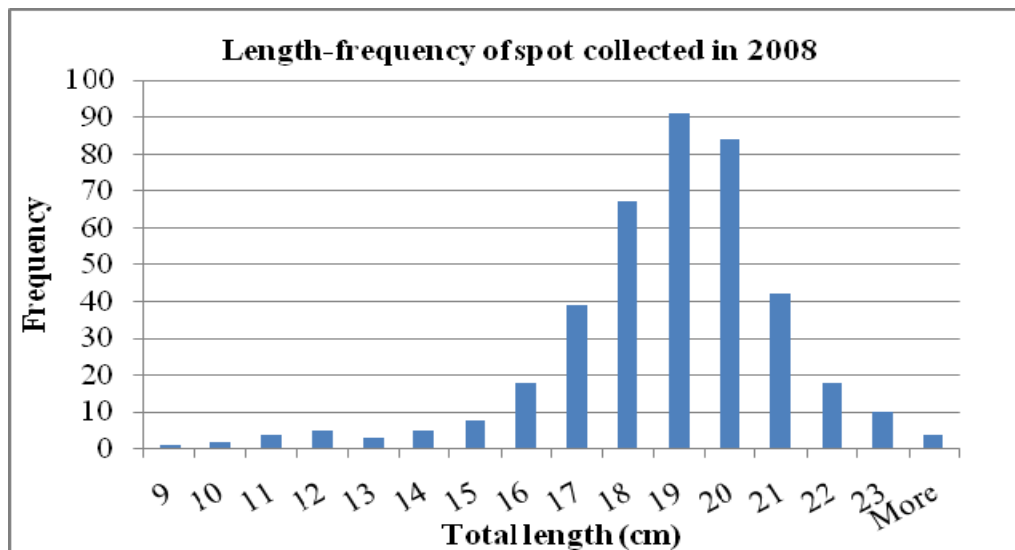


**Figure A7.** Mean CPUE for the three most abundant and most frequently collected elasmobranch species collected in trawls in 2000-2003 and 2008: Atlantic sharpnose (*Rhizoprionodon terraenovae*), bonnethead (*Sphyrna tiburo*) and southern stingray (*Dasyatis americana*). Error bars represent 95% confidence intervals. See Table 7.A2 for results of statistical analyses.

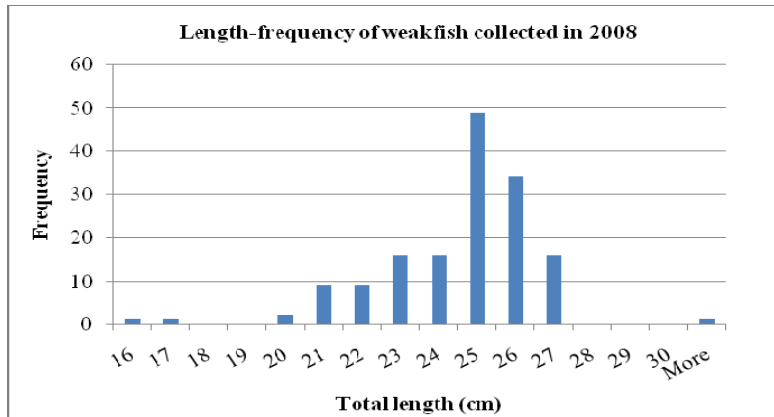
## APPENDIX B – Priority Osteichthyes

**Table B1.** Abundance and size ranges of the 13 species of priority finfish by-catch collected in trawls during 2008. Minimum, maximum, and mean lengths are in centimeters.

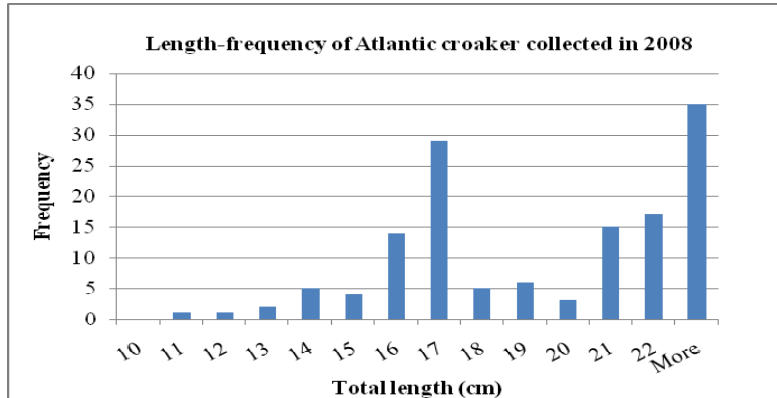
	Osteichthyes	Common Name	# Individ.	Min Size	Max Size	Mean Size
A177	<i>Centropristis striata</i>	black sea bass	12	10	23	17.50
A206	<i>Pomatomus saltatrix</i>	bluefish	8	22	37	26.88
A207	<i>Rachycentron canadum</i>	cobia	8	49	125	80.70
A234	<i>Trachinotus carolinus</i>	pompano	22	32	35	33.50
A263	<i>Archosargus probatocephalus</i>	sheepshead	4	24	25	24.50
A284	<i>Leiostomus xanthurus</i>	spot	409	9	24	18.78
A285	<i>Menticirrhus americanus</i>	southern kingfish	70	16	36	25.69
A288	<i>Micropogonius undulatus</i>	Atlantic croaker	138	11	24	19.43
A362	<i>Scomberomorus maculatus</i>	Spanish mackerel	83	4	47	34.12
A413	<i>Paralichthys dentatus</i>	summer flounder	9	18.5	35	25.94
A414	<i>Paralichthys lethostigma</i>	southern flounder	6	30	49	35.80
A278	<i>Cynoscion regalis</i>	weakfish	157	16	31.8	24.55
A489	Elopidae	tarpons	1	130	130	130.00



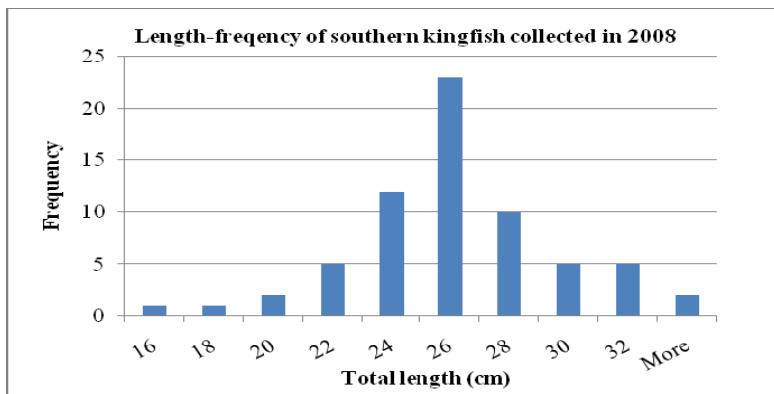
**Figure B1.** Length-frequency of spot (*Leiostomus xanthurus*) caught in trawls during 2008. Lengths are in centimeters.



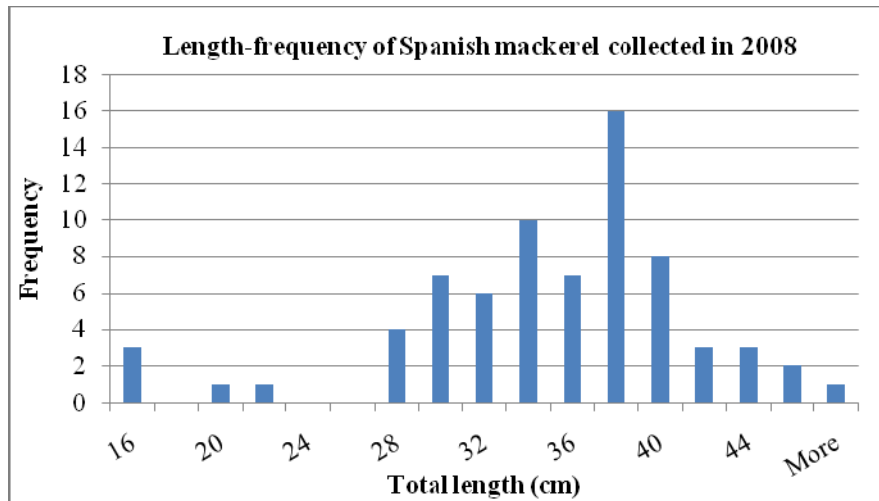
**Figure B2.** Length-frequency of weakfish (*Cynoscion regalis*) caught in trawls during 2008. Lengths are in centimeters



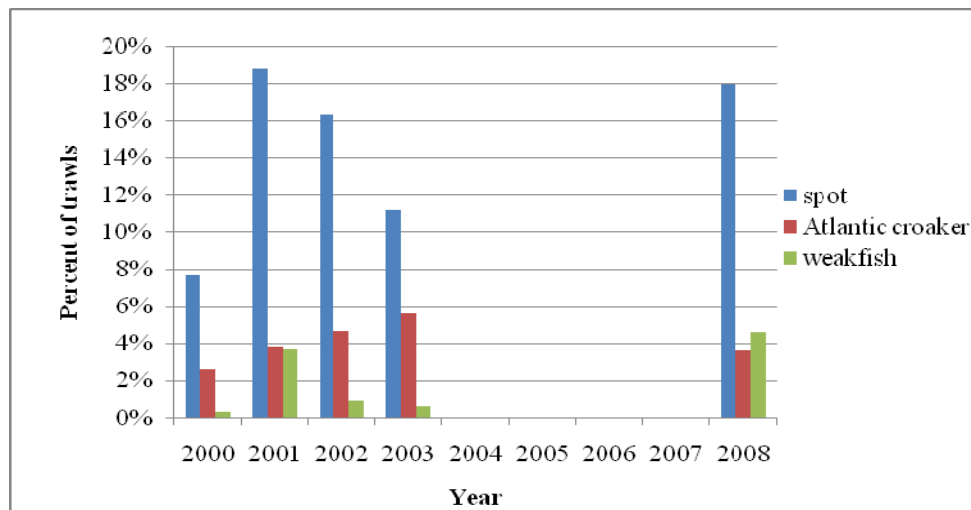
**Figure B3.** Length-frequency of Atlantic croaker (*Micropogonius undulatus*) caught in trawls during 2008. Lengths are in centimeters.



**Figure B4.** Length-frequency of southern kingfish (*Menticirrhus americanus*) caught in trawls during 2008. Lengths are in centimeters.



**Figure B5.** Length-frequency of Spanish mackerel (*Scomberomorus maculatus*) caught in trawls during 2008. Lengths are in centimeters.



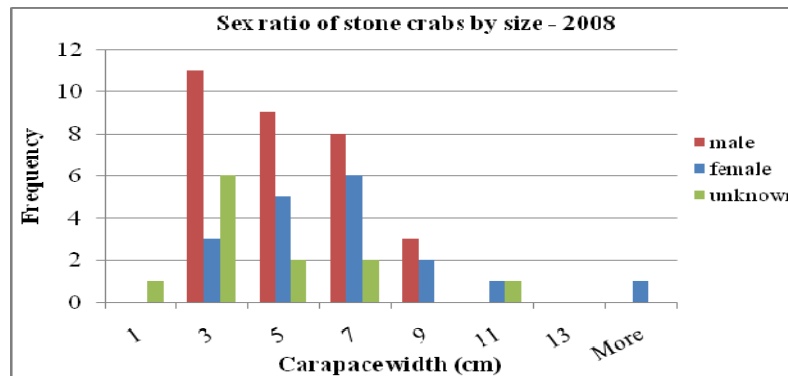
**Figure B6.** Frequency of occurrence of trawls with the three most abundant finfish species during 2000-2003 and 2008: spot (*Leiostomus xanthurus*), Atlantic croaker (*Micropogonius undulatus*) and weakfish (*Cynoscion regalis*). Spot and weakfish were highly significant among years (chi-square;  $p < 0.001$  and  $p < 0.001$ , respectively).



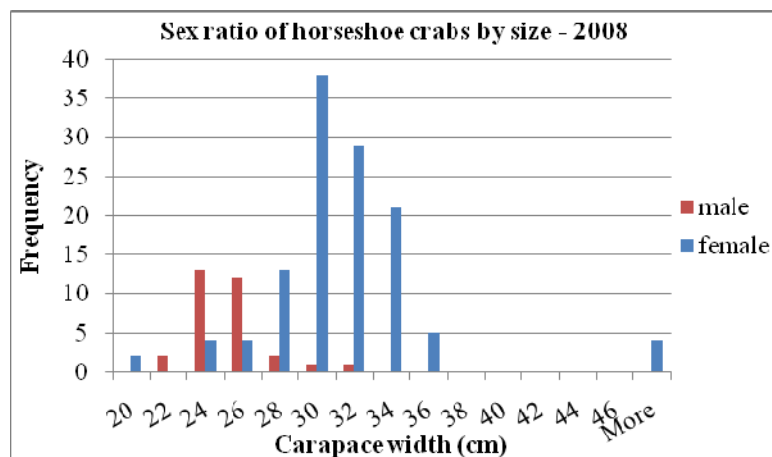
## APPENDIX C – Priority Invertebrates

**Table C1.** Abundance, size ranges, and sex ratios of priority invertebrate by-catch collected in trawls during 2008. Minimum, maximum, and mean lengths are in centimeters. Sex ratios and size-sex distributions are reported only for species with data recorded for  $\geq 25$  individuals.

	Invertebrates	Common Name	# Individ.	Min Size	Max Size	Mean Size	Sex Ratio (M : F : U)
D003	<i>Penaeus aztecus</i>	brown shrimp	22	9	17	12.53	
D005	<i>Penaeus setiferus</i>	white shrimp	12	8	19	15.41	
D130	<i>Callinectes sapidus</i>	blue crab	19	8	18.1	13.71	
D142	<i>Menippe mercenaria</i>	stone crab	70	1	17	5.05	1.6 : 1 : 1
D131	<i>Arenaeus cribrarius</i>	speckled crab	1				
F001	<i>Limulus polyphemus</i>	horseshoe crab	197	19	67	30.05	1 : 3.9 : 1.4
H005	<i>Stomatolophus meleagris</i>	cannonball jelly	6851				



**Figure C1.** Sex ratio by carapace width of stone crabs (*Menippe mercenaria*) caught in trawls during 2008. Carapace widths are in centimeters.



**Figure C2.** Sex ratio by length of horseshoe crabs (*Limulus polyphemus*) caught in trawls during 2008. Carapace widths are in centimeters.